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Effect of nutritional counseling on low-density lipoprotein (LDL) cholesterol among Thai HIV-infected adults receiving antiretroviral therapy

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Effect of nutritional counseling on low-density lipoprotein (LDL) cholesterol among Thai HIV-infected adults receiving antiretroviral therapy

A dissertation submitted in partial satisfaction of the

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in Epidemiology

by

Saipin Chotivichien

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ABSTRACT OF THE DISSERTATION

Effect of nutritional counseling on low-density lipoprotein (LDL) cholesterol among Thai HIV-infected adults receiving antiretroviral therapy

by

Saipin Chotivichien

Doctor of philosophy in Epidemiology

University of California, Los Angeles, 2013

Professor Roger Detels, Chair

Background: Although intensive antiretroviral therapies dramatically reduce mortality rates in HIV-infected patients, significant metabolic complications associated with antiretroviral therapy including dyslipidemia have been increasingly reported. Objectives of this study are to determine 1) an effect of individual nutritional counseling on dyslipidemia, particularly LDL-C, among Thai HIV-infected adults with dyslipidemia who were not currently taking lipid-lowering medication 2) predictors of reducing LDL-C in Thai HIV-infected adults receiving a stable of antiretroviral regimen for at least 3 months.

Methods: We conducted a randomized, 24-week study in HIV-infected patients who were on antiretroviral therapy with dyslipidemia and were eligible to initiate therapeutic lifestyle

changes according to National Cholesterol Education Program (2002). Participants were randomly assigned into two groups. The intervention group received individual counseling with a nutritionist whereas the control group received general diet advice from a physician. A 24-hr recall technique was used to assess dietary intake for both groups at baseline and at week 24. Lipid profile was measured at baseline, at 12 weeks and 24 weeks of follow up. Predictors (measured at baseline) associated with the reduction of LDL-C in HIV-infected patients were assessed for any reduction in LDL-C and for a reduction of at least 10 mg/dL.

Results: Seventy-two patients were randomly assigned. Of these, 62 (86%) completed lipid profile testing and 59 (82%) completed dietary interview. We found a significant difference in mean reduction from baseline of total cholesterol (8% vs 0%) and LDL-C (13% vs 4%) between the intervention group and the control group at week 24. A significant reduction in weight and in carbohydrate intake in the intervention group was observed. Participants who had good level of knowledge of dyslipidemia were more likely to achieve reduction in LDL-C of at least 10 mg/dL at 24 weeks of follow up.

Conclusions: The effectiveness of individual nutritional counseling in improving dyslipidemia, particularly total cholesterol and LDL-C, among Thai HIV-infected adults with dyslipidemia receiving antiretroviral therapy has been demonstrated. To provide a better care for HIV-infected patients receiving antiretroviral therapy with hyperlipidemia, routine HIV/AIDS care with individualized nutritional counseling integration and adequate knowledge of dyslipidemia provision are recommended.

The dissertation of Saipin Chotivichien is approved.

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LIST OF ABBREVIATIONS

AIDS	Acquired immune deficiency syndrome
HIV	Human immunodeficiency virus
ART	Antiretroviral therapy
TLC	Therapeutic lifestyle changes
TC	Total serum cholesterol
LDL	Low-density lipoprotein
HDL	High-density lipoprotein
BMI	Body mass index
SFA	Saturated fatty acid
MUFA	Monounsaturated fatty acid
PUFA	Polyunsaturated fatty acid
PI	Protease inhibitor
NRTI	Nucleoside Reverse Transcriptase Inhibitor
NNRTI	Non-Nucleoside Reverse Transcriptase Inhibitor
MET	Metabolic Equivalent

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PUBLICATIONS

Chotivichien S, Sinawat S, Thammapornpilas P. Sources of daily iodine intake of Thai people 2001. (2002) *Journal of Health Promotion and Environmental Health,* 24, 79-87

Chotivichien S, Jirapongsa C. Investigation of Nosocomial Diarrhea in Nursery ward at Somdej-Prapinklao Hospital, 2002. (2005) *Journal of Health Science*, 14, 345-53.

Chotivichien S. Smart kids with Iodine. (2006) *Journal of clinic*, 22, 273-76.

Chotivichien S, Sinawat S, Thammapornpilas P. Evaluation of growth monitoring and promotion (GMP) surveillance system Thailand 2002. (2006) *Asia-Pacific Journal of Public Health*, 18, 42-45.

Chotivichien S, Krungpanich W, Sinawat S, et al. Monitoring on Iodine Deficiency Disorders (IDDs) status of children in "Mae Fah Luang" Community Learning Centers (CLS), 2005. (2007) *Journal of Health Science* 16, 503-10.

Maleevong K, **Chotivichien S**, Junsawang W, Phonprapai S, et al. Evaluation of integrated growth monitoring by communities in infant and preschool children (0-72 months age). (2008) Journal of Health Education, 31(109): 38-50.

CHAPTER I

Introduction

Background

a. HIV/AIDS and antiretroviral therapy in Thailand

Globally, there were 34.0 million people living with HIV/AIDS at the end of 2011. The number of people newly acquiring HIV infection worldwide is continuing to decline. This is because of the availability and accessibility of antiretroviral therapy. Expanding access to antiretroviral therapy has saved 14 million life-years in low-and middle income countries since 1995 (UNAIDS 2012). Of these, 1.2 million people received antiretroviral therapy for the first time in 2009 alone (Brown, Bao et al. 2010) (UNAIDS 2010). HIV/AIDS is one of top causes of morbidity and mortality among Thais. Between 1984 and September 2012, a total of 276,947 cases attributable to AIDS were reported to the Bureau of Epidemiology, Ministry of Public Health, Thailand (Bureau of Epidemiology). From 2001 to 2011, the incidence of HIV infection among adults 15-49 years old in Thailand decreased by more than 50%. In addition, the number of people dying from AIDS-related causes declined by 25% to 49% from 2005 to 2011.

In Thailand, HIV/AIDS patients can access antiretroviral drugs (ARVs) through the universal coverage benefit scheme for antiviral therapy. By September 2011, there were 247,253 Thai HIV-infected cases treated with ARVs through this universal coverage benefit scheme. The coverage of antiretroviral therapy was approximately 90%. Since 2002, the Government Pharmaceutical Organization (GPO), a state enterprise run by the

Thai Ministry of Public Health, has begun generic production of a number of antiretroviral therapies, including GPO-VIR triple regimen (stavudine, lamivudine, and nevirapine). GPO-VIR is a commonly prescribed antiretroviral therapy in Thailand. A 2-year retrospective cohort reported that approximately one-third (131/325) of HIV-infected patients received GPO-VIR (Ausvapipit J 2005; Tin, Bowonwatanuwong et al. 2005). From the Thai 2010 guidelines for antiretroviral therapy in HIV-1 infected adults and adolescents, a non-nucleoside reverse transcriptase inhibitor (NNRTI)-base regimen containing lamivudine plus tenofovir or zidovudine, not stavudine, is the preferred first regimen for treatment. It is recommended to initiate treatment with ART at CD4 count <350 cells/mm^3 (Sungkanuparph S. 2010).

b. Metabolic complications associated with long-term treatment with antiretroviral therapy

Although receiving antiretroviral therapy has contributed to a decline in deaths among people living with HIV, it comes with long-term metabolic complications such as dyslipidemia, hypertension and diabetes (DM). An association between metabolic complications and long-term treatment of antiviral drugs, particularly protease inhibitors (PI) has been reported in many studies (Samaras, Wand et al. 2007). (Tsiodras, Mantzoros et al. 2000), (Justman, Benning et al. 2003), (Seaberg, Munoz et al. 2005), (Brar, Shuter et al. 2007), (Butt, McGinnis et al. 2009) and (Khaza M 2010). Dyslipidemia, an increase of total serum cholesterol (TC), particularly low-density lipoprotein (LDL) cholesterol, and low levels of high-density lipoprotein (HDL) cholesterol, is one of the cardiovascular disease risk factors and is common in HIV-infected individuals taking highly active

antiretroviral therapy (HAART) (Chuapai, Kiertiburanakul et al. 2007) (Hiransuthikul, Hiransuthikul et al. 2007). An association between dyslipidemia and antiretroviral therapy use has been recognized in several studies. The lipid changes occurred within 3 months and often sooner after initiating antiretroviral therapy and pre-dated the onset of fat redistribution (Green 2002). Results in the Multicenter AIDS Cohort Study (Riddler, Smit et al. 2003) showed that subsequent highly active antiretroviral therapy (HAART) initiation is associated with increases in TC and LDL cholesterol but had little effect on HDL cholesterol level.

Fat Redistribution Syndrome (FRS), characterized by peripheral fat loss often in the cheeks or face or relative central adiposity, frequently accompanied by increased fasting serum cholesterol, triglycerides and insulin resistance, is another pattern of metabolic complication, which also becomes more prevalent in HIV-infected patients receiving antiretroviral therapy, particularly patients who are on protease inhibitor (PI)-containing regimens. A cross-sectional study (Batterham, Garsia et al. 2000) showed that the prevalence of FRS was nearly three-fold higher in those receiving antiretroviral therapy including a PI (59%) than in those receiving antiretroviral therapy who were PI naïve (21%). Likewise, an association of PI use and lipoatrophy has been reported in another cross-sectional study (Ramalho, Goncalves et al. 2011). The study of (Heath, Hogg et al. 2001) showed that increase risk of lipodystrophy (the redistribution of body fat characterized by peripheral fat wasting, increased visceral abdominal fat, breast hypertrophy in women, and enlargement of the dorsocervical fat pad) is associated with the use of PI. An increase in triglyceride and decrease in HDL cholesterol were reported in

a 12-month prospective study (Tremeschin, Sartorelli et al. 2011). Similarly, PI use was reported to be associated with a significant increase in triglyceride and LDL cholesterol in HIV-positive children and adolescents (Contri, Berchielli et al. 2011). Elevation in TC has been associated with PI and/or Non-Nucleoside Reverse Transcriptase Inhibitor (NNRTI) use in other studies (Heath, Hogg et al. 2002) and (Friis-Moller, Weber et al. 2003). Several studies in Thailand have also shown a relationship between antiretroviral therapy and metabolic complications in HIV-infected persons. A recent cross-sectional study showed that 27.5% of 149 HIV-infected patients who visited a hospital in Bangkok were diagnosed as pre-diabetic (Srivanich, Ngarmukos et al. 2010). Metabolic complications such as dyslipidemia, hypertension and DM have been observed in patients who received GPO-VIR as reported in a prospective cohort study. In a 96-week prospective cohort among 140 antiretroviral-naive advanced HIV-infected patients in Thailand who initiate treated with GPO-VIR, dyslipidemia, hypertension and DM were reported in 30%, 9% and 2.3%, respectively (Manosuthi, Tantanathip et al. 2008).

c. Risk factors for metabolic complications in HIV-infected patients receiving antiretroviral drug

Studies on risk factors for metabolic complications in HIV-infected patients receiving antiretroviral drug play an important role in establishing strategies for better HIV care. A study showed that a higher CD4 cell count, older age, white ethnicity and a higher body mass index (BMI) significantly increased the risk of metabolic syndrome among HIV-infected patients (Mondy, Overton et al. 2007). Factors associated with an elevated serum total cholesterol (TC) level were a higher CD4 cell count, older age, lower plasma HIV

RNA levels, clinical signs of lipodystrophy, and longer exposure times to PI and NNRTI (Friis-Moller, Weber et al. 2003; Hiransuthikul, Hiransuthikul et al. 2007). Likewise, an increased intake of total protein, animal protein and trans fat, and reduced soluble fiber consumption contributed to dyslipidemia in HIV-infected subjects on PIs (Shah, Tierney et al. 2005). A cross-sectional study of dyslipidemia in an HIV-positive with antiretroviral therapy (ART)–naïve reported that high triglyceride and low HDL cholesterol levels were significantly associated with low CD4 counts (Armstrong, Liu et al. 2011).

Several factors have been found to be associated with reduction of cholesterol and triglyceride levels among HIV-infected patients with dyslipidemia associated with antiretroviral therapy. Lifestyle modification, particularly diet and exercise, was associated with an 11% reduction in TC level in HIV-infected persons (Henry, Melroe et al. 1998). Dietary counseling and modifying dietary fat intake induced a significant decrease in TC among HIV-infected patients with hypercholesterolemia (Batterham, Brown et al. 2003). A Randomized controlled trial showed that relative to baseline values, patients in dietary advice group consumed less saturated fat with a reduction by 0.07 mmol/L in LDL cholesterol at week 24 of the study (Moyle, Lloyd et al. 2001). Good diet compliance in patients on PIs showed reduction in TC and triglyceride of 13% and 15% at 3 months, whereas only 8% and 7% was observed reduction at 3 months among those who were on PI-sparing regimens (Barrios, Blanco et al. 2002). Individualized dietary advice to modify fat intake (an increase in the polyunsaturated to saturated fat ratio diet) was modestly effective in reducing cholesterol concentration, a 5.3% reduction in blood total cholesterol was shown after at least six months of intervention (Tang, Armitage et al. 1998). A meta-

analysis showed that 2–10 g/d of soluble fiber from oat products, psyllium, pectin, and guar gum each significantly lowered total cholesterol and LDL cholesterol. In contrast, none of the soluble fibers affected triacylglycerols and HDL cholesterol (Brown, Rosner et al. 1999).

There is evidence that diet can ameliorate dyslipidemia in HIV-infected patients. A cross-sectional study showed that a higher intake of polyunsaturated fatty acids (PUFA) was associated with high insulin resistance, whereas those consuming dietary fiber expressed less insulin resistance. Alcohol consumption was associated with both high LDL and HDL cholesterol levels among HIV-infected patients showing fat redistribution, independent of age, sex and PIs use (Hadigan, Jeste et al. 2001). A one-year study with nutritional counseling reported a decrease in serum triglyceride levels by 25% and an increase in HDL cholesterol by 18% in HIV-infected patients with lipodystrophy. None of the participants claimed to exercise regularly. The counseling included behavior modification and encouragement to consume low glycemic-index diet that was also low in saturated fat (<10% total energy) and high in vitamins, fiber (25 g/day), monounsaturated fatty acids (MUFA) (>10% total energy), and PUFA (10% total energy) (Anjos, Pfrimer et al. 2011).

d. Standard care of HIV-infected patients in Thailand

In Thailand, most HIV- positive patients know their HIV status when they are referred to or visiting Bamrasnaradura Hospital. The initiation of antiretroviral therapy is based on patient's symptoms or with a CD4+ T-cell counts < 350 cells/mm^3 following the new Thai 2010 guidelines. Baseline laboratory testing is routinely done in HIV-infected patients

before starting antiretroviral treatment. Their lipid profile is monitored at least once a year. If an abnormal serum lipid profile (TC, LDL, HDL, Triglyceride) is detected in patients without other high risk of cardiovascular disease, physicians will advise patients to control their diet (avoid food with high cholesterol such as oily food, egg yolk, shellfish and dishes with coconut milk). No individual dietary consulting is provided. Then, patients will have their serum lipid profiles repeated in the next 3-4 months. Most physicians usually initiate lipid-lowering medication at this visit if patients still have abnormal lipid profile. However, the decision to provide lipid-lowering medications is made individually by each physician.

e. Relationship among diet intake, plasma cholesterol and coronary heart disease (CHD)

It has been known that an increase of plasma cholesterol level, particularly an increase of plasma LDL cholesterol level contributes to the development of atherosclerosis and enhances the risk for coronary heart disease (CHD) as shown in classic heart hypothesis diagram (Willett 1998). Several papers showed that lowering LDL cholesterol level with lipid-lowering medication reduces the risk of major coronary events and coronary deaths (Downs, Clearfield et al. 1998; Sever, Dahlof et al. 2003; Karalis 2009). It is well known since the 1970s that both type of dietary fatty acids and dietary cholesterol impact plasma cholesterol (Pyorala 1987). The Seven Countries Study (Keys 1970) suggested that dietary saturated fatty acids (SFA) and possibly dietary cholesterol were associated with CHD. Similarly, a prospective analysis of dietary fat and CHD among 80,082 women in 14-year follow-up data from the Nurse's Health Study (Hu, Stampfer et al. 1997) suggested that replacing saturated and trans fat with monounsaturated and polyunsaturated fats was more

effective in preventing coronary heart disease in women than lowering total fat consumption. Several studies also supported that types of fat were more important than total amount of fat in determining the risk of CHD (Hu, Manson et al. 2001). A meta-analysis of prospective studies (Siri-Tarino, Sun et al. 2010) reported that intake of SFA was not associated with an increased risk of CHD (RR 1.07, 95% CI: 0.96, 1.19). However, the study suggested that the specific nutrients used to substitute saturated fat should also be considered. A 19-year follow up study (Shekelle, Shryock et al. 1981) supported the essential in types of fat. The results showed a high dietary intake of cholesterol and low dietary intakes of PUFA were significantly associated with the risk of CHD death. Monounsaturated fatty acids (MUFA), especially oleic acid, was also demonstrated to reduce CHD rate when used as a replacement for saturated fatty acids (Grundy 1987). A pooled analysis of 11 cohort studies (Jakobsen, O'Reilly et al. 2009) suggested that SFA intake should be substituted with PUFA intake rather than MUFA or carbohydrate intake in order to prevent CHD. An overall reduction in risk of coronary events and coronary deaths was reported when substituted PUFA for SFA (Hazard ratio: 0.87; 95% CI: 0.77, 0.97) and (Hazard ratio: 0.74; 95% CI: 0.61, 0.89), respectively. A randomized controlled trial of 48,835 post-menopausal women in the Women's Health Initiative Dietary Modification Trial (Howard, Van Horn et al. 2006) reported that reducing total fat intake to 20% of calories and increasing intakes of vegetables and/or fruits to 5 servings per day significantly reduced LDL cholesterol levels by 3.55 mg/dL, whereas there was no significantly change in levels of HDL cholesterol and triglyceride.

f. Dietary intake of Thai population

The dietary pattern of the Thai population has shifted from a cereal-based and low fat diet to a higher proportion of fats and animal meat diet (Kosulwat 2002). According to the latest Thailand Nutrition Survey (Nutrition Division 2003), males consumed an average of 1,569.1 kilocalories per day, whereas females consumed an average of 1,342.5 kilocalories per day. The ratio of protein: fat: carbohydrate was 13.9: 23.9: 62.1. There is a variety of food in different regions of Thailand. People in the central region have the highest percentage of fat consumption (29.6%), followed by the north (25.4), south (23.3), and northeastern region (19.8). Lipid profiles in healthy Thai adults showed that the prevalence of hyperlipidemia increased with age. An average serum cholesterol of 200$\pm$ 45 mg/dL were reported in the 15-59 years old age group. The highest percentage of hypercholesterolemia (TC $\geq$ 200 mg/dL) was reported in the 50-59 years old age group (47.0%), followed by 40-49 years old age group (35.9%), 30-39 years old age group (31.8%), 20-29 years old age group (23.5%), and 15-19 years old age group (16.3%).

The prevalence of high LDL cholesterol (LDL cholesterol $\geq$ 130 mg/dL) was also increasing with age; 12.8% was reported in the 15-19 years old age group, followed by 22.2%, 24.0%, 29.3% and 36.7% in 20-29 years old age group, 30-39 years old age group, 40-49 years old age group, and 50-59 years old age group, respectively. Thai people still consume less vegetables and fruit than the recommended daily intake level (400 grams per day = 3 servings of vegetables and 2 servings of fruit). According to Bureau Thai Health Survey (2004) (Health System Research Institute (HSRI) 2004) male adults consume an average of 268 grams per day (3.4 servings) of vegetables and fruits, whereas female adults

consume an average of 283 grams per day (3.5 servings). Eighty percent of males and 76% of females consume less vegetables and fruit than the recommended daily intake level.

g. Gaps in the literatures

Little is known about the effect of individual nutritional counseling on lipid profile of HIV-infected patients when receiving ART. Most of the existing published studies were conducted in the western countries and related to lipid-lowering medications. The majority of those existing studies that mentioned the role of lifestyle modification only (diet or/and exercise) on dyslipidemia reduction, menu plans were set for participants during the follow up in order to compare 2 or 3 groups receiving different menu plans. It is not practical to settings like Thailand because diet consumption pattern in Thai different from diet pattern in western people. To date, there are no specific recommendations or guidelines for the management of HIV-associated dyslipidemia. In addition, no nutritional interventions (food-based or supplement) for HIV-infected persons with dyslipidemia exist. In order to help policy makers introduce better strategies to facilitate good care of HIV-infected individuals receiving antiretroviral therapies and make proper decisions on budget allocation for prevention of long-term complications, it is important to identify changes in lifestyle that can reduce known cardiovascular risks, particularly modification of diet and reduction of other factors (e.g., Antiretroviral regimen and exercise) associated with abnormal serum lipid profiles among the HIV-infected.

References

Anjos, E. M., K. Pfrimer, et al. (2011). "Nutritional and metabolic status of HIV-positive patients with lipodystrophy during one year of follow-up." Clinics **66**(3): 407-410.

Armstrong, C., E. Liu, et al. (2011). "Dyslipidemia in an HIV-positive antiretroviral treatment-naive population in Dar es Salaam, Tanzania." Journal of acquired immune deficiency syndromes **57**(2): 141-145.

Ausvapipit J, A. N., Adunyaviwat B, Vipurinat N, Pongchu S, et al. (2005). "Survival rate in HIV/AIDS patients who had CD4 count less than 200 cell/mm3 in Bamrasnaradura Institue." Disease Control Journal **29**(2): 184-192.

Barrios, A., F. Blanco, et al. (2002). "Effect of dietary intervention on highly active antiretroviral therapy-related dyslipemia." AIDS **16**(15): 2079-2081.

Batterham, M. J., D. Brown, et al. (2003). "Modifying dietary fat intake can reduce serum cholesterol in HIV-associated hypercholesterolemia." AIDS **17**(9): 1414-1416.

Batterham, M. J., R. Garsia, et al. (2000). "Dietary intake, serum lipids, insulin resistance and body composition in the era of highly active antiretroviral therapy 'Diet FRS Study'." AIDS **14**(12): 1839-1843.

Brar, I., J. Shuter, et al. (2007). "A comparison of factors associated with prevalent diabetes mellitus among HIV-Infected antiretroviral-naive individuals versus individuals in the National Health and Nutritional Examination Survey cohort." Journal of acquired immune deficiency syndromes **45**(1): 66-71.

Brown, L., B. Rosner, et al. (1999). "Cholesterol-lowering effects of dietary fiber: a meta-analysis." The American journal of clinical nutrition **69**(1): 30-42.

Brown, T., L. Bao, et al. (2010). "Modelling HIV epidemics in the antiretroviral era: the UNAIDS Estimation and Projection package 2009." Sexually transmitted infections **86 Suppl 2**: ii3-10.

Bureau of Epidemiology, D. o. D. C., MOPH, Thailand Situation of AIDS patients in Thailand, date 30 September 2012, retrieved on <http://www.boe.moph.go.th/report.php?cat=19&id=1268>.

Butt, A. A., K. McGinnis, et al. (2009). "HIV infection and the risk of diabetes mellitus." AIDS **23**(10): 1227-1234.

Chuapai, Y., S. Kiertiburanakul, et al. (2007). "Lipodystrophy and dyslipidemia in human immunodeficiency virus-infected Thai patients receiving antiretroviral therapy." Journal of the Medical Association of Thailand = Chotmaihet thangphaet **90**(3): 452-458.

Contri, P. V., E. M. Berchielli, et al. (2011). "Nutritional status and lipid profile of HIV-positive children and adolescents using antiretroviral therapy." Clinics **66**(6): 997-1002.

Downs, J. R., M. Clearfield, et al. (1998). "Primary prevention of acute coronary events with lovastatin in men and women with average cholesterol levels: results of AFCAPS/TexCAPS. Air Force/Texas Coronary Atherosclerosis Prevention Study." JAMA : the journal of the American Medical Association **279**(20): 1615-1622.

Friis-Moller, N., R. Weber, et al. (2003). "Cardiovascular disease risk factors in HIV patients--association with antiretroviral therapy. Results from the DAD study." AIDS **17**(8): 1179-1193.

Green, M. L. (2002). "Evaluation and management of dyslipidemia in patients with HIV infection." Journal of general internal medicine **17**(10): 797-810.

Grundy, S. M. (1987). "Monounsaturated fatty acids, plasma cholesterol, and coronary heart disease." The American journal of clinical nutrition **45**(5 Suppl): 1168-1175.

Hadigan, C., S. Jeste, et al. (2001). "Modifiable dietary habits and their relation to metabolic abnormalities in men and women with human immunodeficiency virus infection and fat redistribution." Clinical infectious diseases : an official publication of the Infectious Diseases Society of America **33**(5): 710-717.

Health System Research Institute (HSRI), M. o. P. H. (2004). The 3rd National Health Examination Survey of Thai people, 2003-2004.

Heath, K. V., R. S. Hogg, et al. (2001). "Lipodystrophy-associated morphological, cholesterol and triglyceride abnormalities in a population-based HIV/AIDS treatment database." AIDS **15**(2): 231-239.

Heath, K. V., R. S. Hogg, et al. (2002). "Antiretroviral treatment patterns and incident HIV-associated morphologic and lipid abnormalities in a population-based chort." Journal of acquired immune deficiency syndromes **30**(4): 440-447.

Henry, K., H. Melroe, et al. (1998). "Atorvastatin and gemfibrozil for protease-inhibitor-related lipid abnormalities." Lancet **352**(9133): 1031-1032.

Hiransuthikul, N., P. Hiransuthikul, et al. (2007). "Lipid profiles of Thai adult HIV-infected patients receiving protease inhibitors." The Southeast Asian journal of tropical medicine and public health **38**(1): 69-77.

Howard, B. V., L. Van Horn, et al. (2006). "Low-fat dietary pattern and risk of cardiovascular disease: the Women's Health Initiative Randomized Controlled

Dietary Modification Trial." JAMA : the journal of the American Medical Association 295(6): 655-666.

Hu, F. B., J. E. Manson, et al. (2001). "Types of dietary fat and risk of coronary heart disease: a critical review." Journal of the American College of Nutrition 20(1): 5-19.

Hu, F. B., M. J. Stampfer, et al. (1997). "Dietary fat intake and the risk of coronary heart disease in women." The New England journal of medicine 337(21): 1491-1499.

Jakobsen, M. U., E. J. O'Reilly, et al. (2009). "Major types of dietary fat and risk of coronary heart disease: a pooled analysis of 11 cohort studies." The American journal of clinical nutrition **89**(5): 1425-1432.

Justman, J. E., L. Benning, et al. (2003). "Protease inhibitor use and the incidence of diabetes mellitus in a large cohort of HIV-infected women." Journal of acquired immune deficiency syndromes **32**(3): 298-302.

Karalis, D. G. (2009). "Intensive lowering of low-density lipoprotein cholesterol levels for primary prevention of coronary artery disease." Mayo Clinic proceedings. Mayo Clinic **84**(4): 345-352.

Keys, A. (1970). "Coronary heart disease in seven countries." Circulation **41**: I-1.

Khaza M, P. K. L., Estari M. (2010). "The effects of protease inhibitors on metabolic complications and lipodystrophy in human immunodeficiency virus-infected patients." American Medical Journal **1**(2): 97-102.

Kongkachuichai, R., R. Charoensiri, et al. (2010). "Carotenoid, flavonoid profiles and dietary fiber contents of fruits commonly consumed in Thailand." International journal of food sciences and nutrition **61**(5): 536-548.

Kosulwat, V. (2002). "The nutrition and health transition in Thailand." Public health nutrition **5**(1A): 183-189.

Manosuthi, W., P. Tantanathip, et al. (2008). "Durability of stavudine, lamivudine and nevirapine among advanced HIV-1 infected patients with/without prior co-administration of rifampicin: a 144-week prospective study." BMC infectious diseases **8**: 136.

Mondy, K., E. T. Overton, et al. (2007). "Metabolic syndrome in HIV-infected patients from an urban, midwestern US outpatient population." Clinical infectious diseases : an official publication of the Infectious Diseases Society of America **44**(5): 726-734.

Moyle, G. J., M. Lloyd, et al. (2001). "Dietary advice with or without pravastatin for the management of hypercholesterolaemia associated with protease inhibitor therapy." AIDS **15**(12): 1503-1508.

Nutrition Division, D. o. H., Ministry of Public Health (2003). "The 5th National Nutrition Survey of Thailand. Bankokok: Express Transportation Organization of Thailand Printing, 2006."

Pyorala, K. (1987). "Dietary cholesterol in relation to plasma cholesterol and coronary heart disease." The American journal of clinical nutrition **45**(5 Suppl): 1176-1184.

Ramalho, L. C., E. M. Goncalves, et al. (2011). "Abnormalities in body composition and nutritional status in HIV-infected children and adolescents on antiretroviral therapy." International journal of STD & AIDS **22**(8): 453-456.

Samaras, K., H. Wand, et al. (2007). "Prevalence of metabolic syndrome in HIV-infected patients receiving highly active antiretroviral therapy using International Diabetes

Foundation and Adult Treatment Panel III criteria: associations with insulin resistance, disturbed body fat compartmentalization, elevated C-reactive protein, and [corrected] hypoadiponectinemia." Diabetes care **30**(1): 113-119.

Seaberg, E. C., A. Munoz, et al. (2005). "Association between highly active antiretroviral therapy and hypertension in a large cohort of men followed from 1984 to 2003." AIDS **19**(9): 953-960.

Sever, P. S., B. Dahlof, et al. (2003). "Prevention of coronary and stroke events with atorvastatin in hypertensive patients who have average or lower-than-average cholesterol concentrations, in the Anglo-Scandinavian Cardiac Outcomes Trial--Lipid Lowering Arm (ASCOT-LLA): a multicentre randomised controlled trial." Lancet **361**(9364): 1149-1158.

Shah, M., K. Tierney, et al. (2005). "The role of diet, exercise and smoking in dyslipidaemia in HIV-infected patients with lipodystrophy." HIV medicine **6**(4): 291-298.

Shekelle, R. B., A. M. Shryock, et al. (1981). "Diet, serum cholesterol, and death from coronary heart disease. The Western Electric study." The New England journal of medicine **304**(2): 65-70.

Siri-Tarino, P. W., Q. Sun, et al. (2010). "Meta-analysis of prospective cohort studies evaluating the association of saturated fat with cardiovascular disease." The American journal of clinical nutrition **91**(3): 535-546.

Srivanich, N., C. Ngarmukos, et al. (2010). "Prevalence of and risk factors for pre-diabetes in HIV-1-infected patients in Bangkok, Thailand." Journal of the International Association of Physicians in AIDS Care **9**(6): 358-361.

Sungkanuparph S., T. W., Utaipiboon C., Chasombat S., Bhakeecheep S., Leechawengwongs M., et al. (2010). "Thai national guidelines for antiretroviral therapy in HIV-1 infected adults and adolescents 2010." Asian Biomedicine **4**(4): 515-528.

Tang, J. L., J. M. Armitage, et al. (1998). "Systematic review of dietary intervention trials to lower blood total cholesterol in free-living subjects." BMJ **316**(7139): 1213-1220.

Tin, E. E., C. Bowonwatanuwong, et al. (2005). "The efficacy and adverse effects of GPO-VIR (stavudine+lamivudine+nevirapine) in treatment-naive adult HIV patients." The Southeast Asian journal of tropical medicine and public health **36**(2): 362-369.

Tremeschin, M. H., D. S. Sartorelli, et al. (2011). "Nutritional assessment and lipid profile in HIV-infected children and adolescents treated with highly active antiretroviral therapy." Revista da Sociedade Brasileira de Medicina Tropical **44**(3): 274-281.

Tsiodras, S., C. Mantzoros, et al. (2000). "Effects of protease inhibitors on hyperglycemia, hyperlipidemia, and lipodystrophy: a 5-year cohort study." Archives of internal medicine **160**(13): 2050-2056.

UNAIDS (2010). Global Report: UNAIDS report on the global AIDS epidemic 2010. Retrieved on http://www.unaids.org/globalreport/global_report.htm.

UNAIDS (2012). "Global Report: UNAIDS report on the global AIDS epidemic 2012. Retrieved on http://www.unaids.org/en/resources/campaigns/20121120_globalreport2012/globalreport/."

Willett, W. (1998). Nutritional Epidemiology. New York, Oxford University Press.

CHAPTER II

Objectives and Methods

I. OBJECTIVES

1. To determine if nutritional counseling will reduce dyslipidemia, particularly LDL cholesterol

2. To identify predictors of reducing LDL cholesterol in HIV-infected patients who have started antiretroviral drug therapy and used the same regimen for at least 3 months

II. METHODS

a. Study site

The study was conducted at Bamrasnaradura Hospital, a 300-bed tertiary care HIV referral center located in Nonthaburi province, Thailand. The Ministry of Public Health (MOPH) nominated Bamrasnaradura Hospital as the National Clinical Reference and Training Centre for HIV/ AIDS in Thailand in 1987. The Bamrasnaradura Hospital treats approximately 40,000 HIV positive outpatients and approximately 2,100 inpatients a year. HIV clinic opens three days per week with 30 – 50 patients per day.

b. Study population

i. Sample size

The sample size is calculated using "Sample Size Calculator" by the Emory University. Using the sample size formula below (Sullivan, Dean et al. 2009) (Kelsey 1996):

$$n_1 = \frac{\left(z_{\alpha/2} + z_{1-\beta}\right)^2 \bar{p}\bar{q}(r+1)}{r\left(p_1 - p_2\right)^2}$$

and $n_2 = r\, n_1$

$$\bar{p} = \frac{p_1 + rp_2}{r+1} \qquad \text{and} \qquad \bar{q} = 1 - \bar{p}$$

Symbol	Definition
n_1	In case-control studies: number of cases; in clinical trial or cross-sectional or cohort studies: number of exposed
n_2	In case-control studies: number of controls; in clinical trial or cross-sectional or cohort studies: number of unexposed
$Z_{\alpha/2}$	Standard normal deviate for two-tailed test based on alpha level (relates to the confidence interval level)
Z_{β}	Standard normal deviate for one-tailed test based on beta level (relates to the power level)
r	In case-control studies: ratio of controls to cases; in clinical trial or cross-sectional or cohort studies: ratio of unexposed to exposed
p_1	In case-control studies: proportion of cases who are exposed; in clinical trial or cross-sectional or cohort studies: proportion of exposed with disease and $q_1 = 1\text{-}p_1$
p_2	In case-control studies: proportion of controls who are exposed; in clinical trial or cross-sectional or cohort studies: proportion of unexposed with disease and $q_2 = 1\text{-}p_2$

To determine if nutritional counseling in HIV-positive individuals will reduce LDL-cholesterol, we will select only HIV positive individuals who have LDL-cholesterol $\geq$100 mg/dL ($\geq$ 2.6 mmol/L) at baseline. The numbers of participants are planned with a 1: 1 randomization. Groups will be assigned by generating two regimens (standard care group and individual counseling with nutritionist group). Assuming the percentage of those in the standard care group (without individual counseling) will develop the outcome of interest

(decrease in LDL-cholesterol level) during the 24 weeks is 35%, whereas the proportion that develops the outcome of interest (decrease in LDL-cholesterol level) during the 24 weeks is 60% in intervention group (with individual counseling). Therefore given alpha level of 0.05 and power of 80%, the sample size is approximate 126 HIV positive persons with high LDL-cholesterol ($\geq$100 mg/dL). We assume about 20% loss to follow up so the final sample size for the randomized controlled trial was 152 HIV positives with high LDL-cholesterol, which was 76 participants in each group.

Due to the difficulty in recruiting eligible participants and limited time frame, the study ended up with a total of 72 participants, of whom 35 and 37 participants were randomized into intervention group and control groups, respectively. Of 72 participants; 62 participants (14% loss to follow up) completed their lipid profile tests (at baseline, week 12 and 24), 64 participants (11% loss to follow up) had their lipid tests only at baseline and week 24, and 59 participants (18% loss to follow up) completed their dietary assessments at 24 weeks of follow up period. Under the same assumption about the percentage of developing outcome of interest in each group mentioned above, the number of participants we had provided power of 50% to detect the predicted difference.

ii. Criteria for inclusion and exclusion

The study population was HIV-infected patients with abnormal LDL cholesterol who were treated with antiretroviral therapy and visited the outpatient department (OPD) at Bamrasnaradura Hospital. Criteria for inclusion in the study included: age between 18 and 65 years old, have started stable antiretroviral therapy for more than 3 months, and were

not currently treated with the lipid-lowering or/and diabetes medication. Subjects were excluded if they had a history of cancers, renal failure, pancreatitis or liver cirrhosis, were pregnant or were illiterate persons who unable to complete the food record.

c. Recruitment of participants

Participants were recruited from HIV-infected patients with abnormal LDL-cholesterol who were on antiretroviral therapy at least 3 months and had visited the outpatient department (OPD) at Bamrasnaradura Hospital. The principal investigator contacted infectious disease physicians who take care of HIV-infected patients. When there was an HIV-infected person with hyperlipidemia above LDL cholesterol goal (Table 1) from the process mentioned above, a recruitment flyer was distributed to the potential participants by a physician. Potential participants were referred to project recruiting staff. Then, the project recruiting staff invited those potential participants to participate the study and screened them with some questions and risk of cardiovascular disease to determine their eligibility (Appendix 1). Before conducting the interview, an investigating team asked eligible participants if they agreed to participate in the study. The requirements and benefits of the study were provided to them. Participants were requested to sign the informed consent form if they agreed to participate the study.

d. Randomization process

Eligible participants who agreed to participate in the study were randomized into two groups, by a research staff, to either: 1) individual counseling with a nutritionist group; or 2) standard care group which no individual counseling with a nutritionist. The

randomization block of size 10 was used to assign the group to those participants in order to balance the numbers in each group. The block of size 10 could create 45 possible permuted blocks, each block contained sequence for 10 patients with five patients in intervention group and five patients in control group. A staff from Ministry of Public Health who did not know any information about participants selected 16 permuted blocks. Then, sequentially numbered, opaque sealed envelopes (SNOSE) method (Doig and Simpson 2005) was used to prepare group enveloped according to 16 permuted blocks mentioned above. These prepared envelopes for each permuted blocks were placed in a plastic box with fitting cover. A research staff who was blinded with information of participants picked the envelope on the top in order to assign the eligible participants who were recruited on that day. The next participant was assigned to the group mentioned in the next top envelope.

e. Dietary intervention

Participants in the intervention group were advised to follow dietary guidelines from the National Cholesterol Education (NCEP) program (2002) as showed below (Table 2):

- Reduction of daily calories from total fat to less than 25 percent
- Reduction of saturated fat to less than 7 percent
- Reduction of cholesterol to less than 200 mg a day
- Increase of polyunsaturated fat up to 10 percent of total calories
- Increase of monounsaturated fat up to 20 percent of total calories
- Consumption of fiber approximately 20-30 g/day
    - 10-25 grams per day of soluble fiber

- Consume protein approximately 15 percent of total calories
- Consume carbohydrate 50-60% of total calories

Table 1. LDL Cholesterol Goals and Cut points for Therapeutic Lifestyle Changes (TLC) and Drug Therapy in Different Risk Categories according to NCEP (2002)

Risk Category	**LDL Goal**	**LDL level**	
		Initiate TLC	**Consider Drug Therapy**
CHD or CHD risk equivalents (10-year risk > 20%, noncoronary atherosclerotic vascular disease, or type 2 DM)	<100 mg/dL (<2.6 mmol/L)	≥100 mg/dL (≥ 2.6 mmol/L)	≥130 mg/dL (≥ 3.4 mmol/L) 100-129 mg/dL (2.6-3.3 mmol/L): drug optional)
2 or more risk factors∗ (10-year risk ≤ 20%)	<130 mg/dL (<3.4 mmol/L)	≥130 mg/dL (≥ 3.4 mmol/L)	*10-year risk of 10-20%:* ≥130 mg/dL (≥ 3.4 mmol/L) *10-year risk <10%:* ≥160 mg/dL (≥ 4.1 mmol/L)
0 or 1 risk factor∗	<160 mg/dL (<4.1 mmol/L)	≥160 mg/dL (≥ 4.1 mmol/L)	≥190 mg/dL (≥ 4.9 mmol/L) 160-189 mg/dL (4.1-4.9 mmol/L): drug optional)

∗Risk factors include cigarette smoking; hypertension; HDL cholesterol level below 40 mg/dL; family history of premature CHD; age (>45 years for men and >55 years for women). Risk factor equivalent: diabetes.

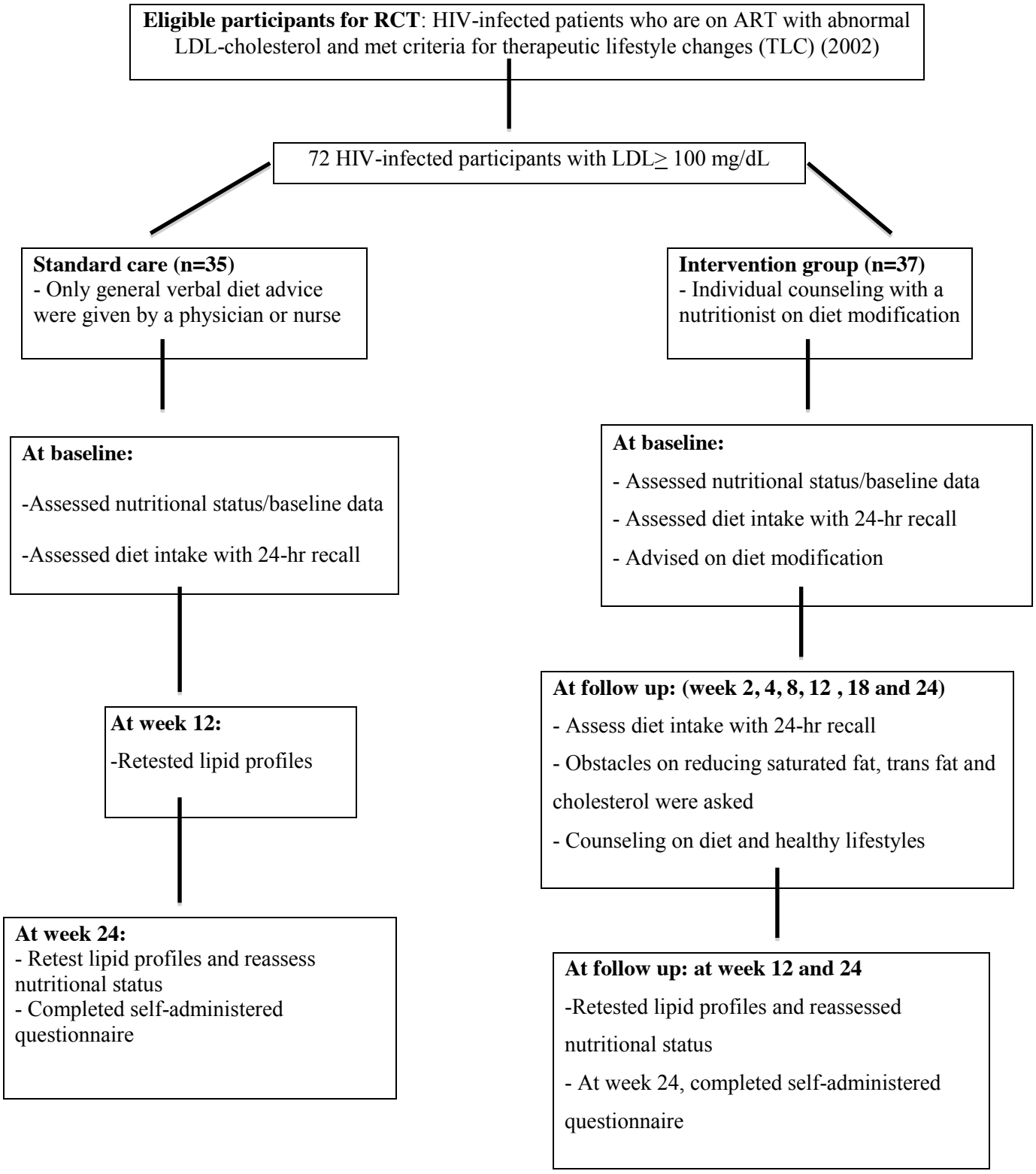

Figure 1. Diagram of study participants' recruitment

Table 2. Approximate and cumulative LDL cholesterol reduction achievable by dietary modification (2002)

Dietary Component	Dietary Change	Approximate LDL-C Reduction
Major		
Saturated fat	<7% of calories	8-10%
Dietary cholesterol	<200 mg/day	3-5%
Weight reduction	Lose 10 lbs	5-8%
Other LDL-lowering options		
Soluble fiber	5-10 g/day	3-5%
Plant sterol/stanol esters	2 g/day	6-15%
Cumulative estimate		20-30%

Table 3. Thai Food-Based Dietary Guidelines (Nutrition Division 2001)

Food group	Measuring Tools	Energy used (Kcal)*		
		1,600	**2,000**	**2,400**
Rice – starchy food	Rice-serving spoons	8	10	12
Vegetable	Rice-serving spoons	6	5	6
Fruit	Portions	4	4	5
Meat	Spoons	6	9	12
Milk	Glass	1	1	1
Oil, sugar and salt	Teaspoon	Eat these items in limited amounts		

*1,600 Kcal: Working women: 25-60 years old and elderly: older than 60 years old
2,000 Kcal: Teenagers and young adults: 14-25 years old, working men 25-60 years old
2,400 Kcal: Those who need more energy such as laborers, farmers, athletes etc.

Thai Food-Based Dietary Guidelines was introduced to participants according to their energy need per day to reach or maintain a healthy weight (Table 3). In addition, participants were advised to stop smoking and reduce alcohol consumption (if any), and encouraged to engage in at least 30 minutes of a moderate-intensity physical activity, such as brisk walking, on most, and preferably all, days of the week.

Measuring Tools: Rice-serving spoon, spoon, portion, and glass/cup

Rice-serving spoon

- Rice: 1 rice-serving spoon equals 60 grams or about 1/2 cup
- Cooked vegetable: 1 rice-serving spoon equals 40 grams or about 1/2 cup

Spoon (=tablespoon)

- Cooked meat: 1 spoon is about 15 grams; 1/2 mackerel or 1/2 egg or 1/4 hard tofu

Portion

- Fruit: Examples of 1 portion of fruit are 1 banana, 1 orange or 4 rambutans. Examples of 1 portion for big fruit: 6-8 pieces of papaya or pineapple or watermelon.

Nutritionists recommended a low saturated fat, high polyunsaturated fat or/and monounsaturated fat and high soluble fiber diet to participants using data from fatty acid and cholesterol in Thai foods (Nutrition Division 2002), and use food sources of soluble fiber from National Heart Lung and Blood Institute ((NHLBI) 2006) on lowering cholesterol with Therapeutic Lifestyle Changes (TLC) ((NHLBI) 2006), a study of dietary fiber contents of fruits commonly consumed in Thailand (Kongkachuichai, Charoensiri et al. 2010) and dietary fiber contents in Thai food from Bureau of Nutrition, Ministry of

Public Health (Bureau of Nutrition 2012).

Contents of nutritional counseling included the advice on reducing saturated fat, trans fat, and cholesterol and increasing soluble fiber.

1. Reduce consumption of diet that is major sources of saturated fatty acids. These diet are shown as following: (Aim: reduce to <7% of total energy).

1.1 High-fat dairy products such as whole milk, cheese, butter, ice cream, chocolate and cream.

1.2 High-fat meats such as beef, pork, bacon and sausage

1.3 Coconut products

1.4 Tropical oils such as palm oil, coconut oil, and palm kernel oil

1.5 Baked products and mixed dishes containing dairy fats, shortening, and tropical oils.

2. Avoid consumption of diet that contains trans fatty acid. These diet are shown as following: (Aim: intakes should be kept low)

2.1 Products made from partially hydrogenated oils such as baked products including crackers, cookies, doughnut, breads, and products like French fries or chicken fried in hydrogenated shortening.

3. Reduce consumption of diet that is rich in cholesterol, these diet are shown as following: (Aim: reduce to <200 mg a day).

3.1 Egg yolk, high-fat meat, and high-fat dairy products

3.2 Entrails

3.3 Poultry

3.4 Shellfish

4. Substitution of polyunsaturated fatty acid (PUFA) for saturated fatty acid. The sources of PUFA are shown as following: (Aim: increase PUFA up to 10% of total calories)

4.1 Liquid vegetable oils

4.2 Fish oils, corn, soybean, safflower, sunflower, and cottonseed oils (these food items are rich in omega-3)

4.3 Semi-liquid margarines and other margarines low in trans fatty acids

5. Increase consumption of dietary patterns that are rich in monounsaturated fatty acids (MUFA). The sources of MUFA are shown as following: (Aim: increase MUFA up to 20% of total calories)

5.1 Plant oils: olive and canola oil

5.2 Nuts: peanuts, cashews, almonds, and most other nuts

5.3 Whole grains such as brown rice, oat and wheat

5.4 Avocado

5.5 Olive

6. Increasing soluble fiber in the diet. The sources of soluble fiber are shown in Table 4 and Table 5: (Aim: 10–25 grams per day of soluble fiber)

7. Recommend healthy lifestyles

7.1 Physical activity

- Encourage at least 30 minutes of a moderate-intensity physical activity, such as brisk walking, on most, and preferably all, days of the week.

7.2 Alcohol consumption

7.2.1 For people who do not drink alcohol, advise not to initiate alcohol consumption

7.2.2 If drink; (A drink is defined as 5 ounces of wine, 12 ounces of beer, or one and

a half ounces of 80 proof whiskey.

- Men: No more than two drinks per day
- Women: No more than one drink per day

7.3 Stop cigarette smoking (if currently smoke)

Table 4 Food sources of soluble fiber ((NHLBI) 2006)

Food Source	Soluble Fiber (g)	Total Fiber (g)
Cereal Grains (1/2 cup cooked)		
Barley	1	4
Oatmeal	1	2
Oat bran	1	3
Seeds: Psyllium Seeds, Ground (1 Tbsp.)	5	6
Fruit (1 medium fruit)		
Apple	1	4
Bananas	1	3
Blackberries (1/2 cup)	1	4
Citrus Fruit (orange, grape fruit)	2	2-3
Nectarines	1	2
Peaches	1	2
Pears	2	4
Plums	1	1.5
Prunes (1/4 cup)	1.5	3
Legumes (1/2 cup cooked)		

Bean		
- Black Beans	2	5.5
- Kidney Beans	3	6
- Lima Beans	3.5	6.5
- Lentils (yellow, green, orange)	1	8
Peas		
- Chick Peas	1	6
- Black Eyed Peas	1	5.5
Vegetables (1/2 cup cooked)		
- Broccoli	1	1.5
- Brussels Sprouts	3	4.5
- Carrots	1	2.5

Table 5 Dietary fiber contents in Thai food (Thai Bureau of Nutrition 2012)

Food Source	Soluble Fiber (g/100 g)	Total Fiber (g/100 g)
Cereal Grains and Legumes		
Oat	3.39	5.92
Brown rice	0.58	1.67
Black glutinous rice	0.94	3.91
Job's tears	0.33	3.17
Soybean	1.27	11.76
Black lentil	1.32	11.45

Kidney bean	3.34	19.16
Fruit		
Pineapple	0.23	0.83
Barracuda mango	0.61	1.00
Rambutan, Rong-rean variety	0.82	1.41
Dragon fruit, white	0.26	1.24
Mushroom		
Shitake mushroom	3.83	31.33
Sarjor-caja mushroom	0.21	4.47
Straw mushroom	0.18	2.09
Vegetables		
Pea eggplant	1.64	9.27
Water morning glory	0.31	3.02
Winged Bean	0.19	2.91
Yard long bean	0.11	2.88
Egg plant	1.26	2.59
Cauliflower	0.84	2.53
String bean	0.56	1.90
Sugar snap pea	0.49	1.78
Bamboo Shoot, pai-tong	0.15	1.76
Bitter gourd	0.64	1.76
Holy basil	0.29	1.69
Cabbage	0.48	1.53

Mung Bean sprouts	0.31	1.05
Luffa, Ridge gourd	0.30	1.19
Chinese cabbage	0.38	1.18
Cherry tomato	0.25	0.86
Peach tomato	0.24	0.82

Participants were advised on how to limiting saturated fat, trans fat and cholesterol in their cooking. Nutritionist emphasized on the following points ((NHLBI) 2006);

Meat, Poultry, and Fish

- Choose fish, poultry, and lean cuts of meat. Trim the fat from meats; remove the skin and fat from chicken. Keep portion sizes as what recommended in Thai Food-Based Dietary Guidelines (Nutrition Division 2001)
- Broil, bake, roast, or poach instead of frying. When you do fry, use a nonstick pan and a nonstick cooking spray or a very small amount of oil or margarine. Use plant oils, such as olive, soybean, or canola oil, instead of tropical oils, such as palm oil, coconut oil, and palm kernel oil.
- Cut down on sausage, bacon, and processed, high-fat cold cuts.

Milk Products and Eggs

- Instead of whole milk or cream, use fat-free or low-fat milk.
- Replace ice cream with sorbet, sherbet, and fat-free or low-fat frozen yogurt. Keep portion sizes moderate.
- Limit the number of egg yolks (have no more than four a week) ((NHLBI) 2008).
- If use margarines, choose soft margarines (liquid or tub types) that contain little or

no trans fat.

Grains and Grain Products

- Eat foods with lots fiber. Choose whole grain such as whole-grain breads, pasta, and cereals, as well as brown rice.

Sauces, Soups, and Casseroles

- After making sauces or soups, cool them in the refrigerator and skim the fat from the top. Do the same with canned soups.
- Thicken a low-fat sauce with cornstarch or flour.
- Make main dishes with whole-grain pasta, brown rice, or dry peas and beans. If add meat, use small pieces for flavoring rather than as the main ingredient.

For intervention group, a Thai manual on nutritional promotion for HIV-infected patients with dyslipidemia (Chotivichien S 2012) was distributed to participants at baseline. The contents in the manual included example of menu plans with calorie calculation for each menu and the information described above.

Schedule for nutritional counseling sessions

Session 1 (week 0): Baseline assessment, including demographic data, anthropometric measurement, lifestyle habits (physical activity and alcohol consumption) and diet assessment with 24-hr recall.

Session 2 (week 2): Dietary assessment with 24-hr recall. Initiation of therapeutic lifestyle change (TLC) diet. Participants were advised on diet patterns that are reducing on saturated fat, trans fat and cholesterol.

Session 3 (week 4): Dietary assessment with 24-hr recall. Identify problems and obstacles to TLC dietary adherence. Encourage continuing lifestyle changes.

Session 4 (week 8): Dietary assessment with 24-hr recall. Intensifying the TLC diets for LDL cholesterol lowering. Encouraged healthy eating focus on more vigorous reduction in saturated fats and cholesterol and increase soluble fiber.

Session 5 (week 12): Dietary assessment with 24-hr recall. Reassess fasting serum lipid profiles. A decision was made whether the participant should be consider adding drug therapy if the LDL cholesterol goal has not been achieved. Initiate lifestyle therapies such as weight control and increased physical activity.

Session 6 (week 18): Dietary assessment with 24-hr recall. Intensifying the TLC diets for LDL cholesterol lowering and continuing lifestyle changes encouragement

Session 7 (week 24): Reassess diet intake (with 24-hr recall), nutritional status and fasting serum lipid profiles.

f. Measurements

The study involved several dietary assessments and two interviews with self-administered questionnaire at baseline and 24 weeks, as well as follow-up serum lipid profiles at 12 weeks and 24 weeks. Participants in the intervention group received individual counseling and asked to come for six more visits to the hospital in the 24-week period, including 2, 4, 8, 12, 18, and 24 weeks. Participants in the control group received no individual counseling by a nutritionist and asked to come for two more visits at 12 and 24 weeks for testing their serum lipid profiles. At each visit, research staff measured participants' blood pressure, weight, height, waist and hip circumferences. The interview, including weighing and

measurements, took about 45 - 60 minutes. Self-administered questionnaire is illustrated in Appendix 2. Detailed information regarding measurements of outcome variables is provided in the following chapters.

Exposures and or covariates

i. Demographic factors

Age: defined as the respondents' age in years at the time of the first interview.

Gender: defined as the respondents' biological gender (male or female).

Educational level: defined as the highest educational level obtained: illiterate, elementary schools (6 years), secondary schools (6 years); 3 years of lower- and 3 years of upper-secondary levels including vocational upper secondary schools, university including undergraduate (4-6 years) and postgraduate (2-5 years).

Occupation: defined as a respondents' usual or principal work in earning a living.

Marital status: Marital status at the time of the first interview is categorized as *never married*, *married* (living with a spouse and having a marriage certificate), *live-in* (living with the spouse without marriage certificate), *separated* (separate or divorce from the spouse) and *widowed* (spouse died and respondent has not married again).

Alcohol use: Frequency of drinking will be defined as; ≤1 drink/ month, 2-3 drinks /month, 1-2 drink(s)/ week, 3-4 drinks/week, almost every day and daily.

Physical activity:

Inactive – report not engaging in any of the following activities during the previous month: walking, jogging, bike riding, swimming, aerobics, dancing, calisthenics, gardening, lifting weights, or other physical activity outside of their occupation.

Active - report engaging in any of the following activities during the previous month: walking, jogging, bike riding, swimming, aerobics, dancing, calisthenics, gardening, lifting weights, or other physical activity outside of their occupation.

ii. Anthropometric measurements

Body mass index (BMI) computed as body weight (kgs)/[height (meters)]^2: categories were defined as; ([Anon] and Who 2004)

BMI <18.5 kg/m2 (underweight), 18·5–24·9 kg/ m^2 (normal range), 25–29·9 kg/m^2 (preobese), $\geq$ 30 kg/m^2 (obesity)

Waist circumference was measured at the narrowest circumference of the torso. The cut-off points were chosen according to the International Diabetes Federation cut-off points for South Asians, Chinese and Japanese populations (Alberti, Zimmet et al. 2007).

Men	> 90 cm
Women	> 80 cm

Hip circumference was measured over the greater trochanters of the hips with the subject standing. The unit is centimeters.

Waist—hip ratio (WHO 2008)

Abdominal obesity is defined as waist-hip ratio $\geq$ 0.90 for males and $\geq$ 0.85 for females.

iii. Risk of cardiovascular diseases

Hypertension was defined as blood pressure exceeding 140/90 mmHg.

Smoking: Current smoking and never smoking/ex-smoking

Family history of chronic diseases related to cardiovascular disease risks

-Dyslipidemia/Diabetes/hypertension/Obesity/Cardiovascular diseases (stoke, myocardial infarction, etc.)

Family history of cardiovascular disease was defined as myocardial infarction in a first-degree male relative aged <55 years or a female relative aged <65 years.

iv. Antiretroviral therapy history and route of HIV acquisition

Route of HIV acquisition defined as heterosexual, homosexual, bisexual, intravenous drug use, blood transfusion, tattoo and unknown.

Duration of being known as HIV positive: continuous variable (years)

CD4 count: was collected from medical records of HIV positives as done by the Bamrasnaradura Hospital. This collection was approved by the patients and the Ethics Board of Bamrasnaradura hospital.

Type of Anti-retroviral Therapy (ART)

Class	Generic Name	Abbreviation
NRTIs (Nucleoside/ Nucleotide Reverse Transcriptase Inhibitors)		
	Lamivudine	3TC
	Zidovudine	AZT or ZDV
	Stavudine	D4T
	Abacavir	ABC
	Didanosine	ddl
	Emtricitabine	FTC
Non-Nucleoside Reverse Transcriptase Inhibitors (NNRTIs)		
	Nevirapine	NVP

	Efavirenz	EFV
	Delaviridine	DLV
	Etravirine	ETV
Protease Inhibitors (PIs)		
	Indinavir	IDV
	Nelfinavir	NFV
	Fosamprenavir	FPV
	Saquinavir	SQV
	Tipranavir	TPV
	Ritonavir	RTV
	Davunavir	DRV
	Atazanavir	ATV
	Lopinavir + ritonavir	LPV/r

v. Knowledge of dyslipidemia

Patients answered ten True-False questions in a self-administered questionnaire to determine their level of knowledge regarding dyslipidemia. The variable was collected as the number of correct answers.

- Good = 8-10 correct answers
- Fair = 6-7 correct answers
- Poor = 0-5 correct answers

vi. Diet pattern

Dietary intake was assessed using a single 24-hr recall by trained nutritionists

g. Data management

Participants completed the self-administered questionnaire in a private room in the hospital. If there was any question that the participants do not understand, they were encouraged to ask trained interviewers, who stayed in the next room. When each participant completed the questionnaire, they were asked to return the questionnaire back to the interviewer in the next room. The interviewer browsed the questionnaire quickly with the participant's permission to check completeness and logical consistency of the answers. The missing and inconsistent responses were investigated for the reasons. The interviewers clarified those questions to the participant if the reason was due to the difficulty of understanding the questions by the participant. The interviewer reassured the participants regarding the confidentiality of the answers and confidentiality of the interview if the reason was due to their worry about privacy. Information on treatment records at the hospital regarding lipid profile, CD4+ and type of ART was reviewed by the principal investigator (PI) after received permission from the participants. Completeness and logical consistency of the answers in the questionnaires were repeated by PI at the end of session when combined data from participants on that day. If missing values or mistakes in filling the questionnaires were observed, the PI would inform the interviewers and discuss the reasons leadings to the mistakes and how to prevent and correct them. The problems were discussed during the meetings that were held once a week. Data was stored in PI's computer and a research staff's computer. A back up of data was made and stored in the computer of PI.

h. Data analysis

The datasets were backed up and protected by a password. Original questionnaires and all participants' data using in this study were locked in a safe place. Only PI and another research staff had the key to access to the stored file. Data was analyzed by using SAS (version 9.13). Details of the study are described in the following chapters.

References

(2002). "Third Report of the National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel III) final report." Circulation 106(25): 3143-3421.

(NHLBI), U. S. D. o. H. a. H. S. N. H. L. a. B. I. (2006). Your Guide to Lowering Cholesterol with Therapeutic Lifestyle Changes (TLC).

(NHLBI), U. S. D. o. H. a. H. S. N. H. L. a. B. I. (2008). Healthy Hearts, Healthy Homes - Do You Know Your Cholesterol Levels?

[Anon] and Who (2004). "Appropriate body-mass index in Asian populations and its implications for policy and intervention strategies. (vol 363, pg 157, 2004)." Lancet 363(9412): 902-902.

Alberti, K. G., P. Zimmet, et al. (2007). "International Diabetes Federation: a consensus on Type 2 diabetes prevention." Diabetic medicine : a journal of the British Diabetic Association 24(5): 451-463.

Bureau of Nutrition, D. o. H., Ministry of Public Health (2012). "Dietary Fiber Contents in Thai Foods."

Chotivichien S, P. S., Thongbuasirilai N, Srisura W, Sinawat S, Saiwongse N, Viriyautsahakul N. (2012). "A manual on nutritional promotion for HIV-infected patients with dyslipidemia (in Thai)." Bangkok: A.V. Progressive Press, 2012. Print. .

Doig, G. S. and F. Simpson (2005). "Randomization and allocation concealment: a practical guide for researchers." Journal of critical care 20(2): 187-191; discussion 191-183.

Kelsey, J. L. (1996). Methods in observational epidemiology. New York ; Oxford, Oxford University Press.

Nutrition Division, D. o. H., Ministry of Public Health (2001). "Manual Nutrition Flag: healthy eating for Thais."

Nutrition Division, D. o. H., Ministry of Public Health (2002). "Fatty acids composition and cholesterol in Thai foods."

Sullivan, K. M., A. Dean, et al. (2009). "OpenEpi: a web-based epidemiologic and statistical calculator for public health." Public health reports 124(3): 471-474.

WHO (2008). Waist Circumference and Waist-Hip Ratio: Report of a WHO Expert Consultation. Geneva, 8-11 December 2008.

CHAPTER III

Effect of nutritional counseling on low-density lipoprotein (LDL) cholesterol among Thai HIV-infected adults receiving antiretroviral therapy

Abstract

Background: HIV-infected patients receiving antiretroviral therapy encounter the metabolic syndrome, including dyslipidemia. In this study, we determined whether individual nutritional counseling reduced dyslipidemia, particularly LDL cholesterol, among HIV-infected patients with dyslipidemia who were not currently taking lipid lowering medication.

Methods: We conducted a randomized, 24-week study in HIV-infected patients who were on antiretroviral therapy with dyslipidemia and were eligible to initiate therapeutic lifestyle changes (TLC) according to National Cholesterol Education Program. Participants were randomly assigned into two groups. The intervention group received individual counseling with a nutritionist for 7 sessions (at baseline, week 2, 4, 8, 12, 18 and 24) whereas the control group received nutritional counseling only at week 24. A 24-hr recall technique was used to assess dietary intake for both groups at baseline and at week 24. Lipid profile (total cholesterol, LDL, HDL and triglyceride) was measured at baseline, after 12 weeks and 24 weeks of therapy.

Results: Seventy-two patients were randomly assigned and 62 (86%) participants completed their lipid profile test. After 12 weeks of follow-up, significant changes in total cholesterol (-14.4 $\pm$ 4.6 mg/dL, P =0.002), LDL cholesterol (-13.7 $\pm$ 4.1 mg/dL, P =0.001) and triglyceride (-30.4 $\pm$ 13.8 mg/dL, P =0.03) were observed in the intervention group. A significant reduction in LDL cholesterol was also observed in the control group (-7.7 $\pm$ 3.8 mg/dL, P =0.04). However, there were no significant differences of the changes of mean lipid levels between groups at 12 weeks of follow up. After 24 weeks, compared with the control group, participants randomly assigned in the intervention group demonstrated significant decreases in serum in total cholesterol (-19.0 $\pm$ 4.6 versus 0.2 $\pm$ 4.3 mg/dL, P = 0.003) and LDL cholesterol (-21.5 $\pm$ 4.1 versus -6.8 $\pm$ 3.8 mg/dL, P = 0.009). However, there were no significant changes in HDL cholesterol or triglyceride level in either group.

Conclusions: The data demonstrated that sessions of individual nutritional counseling significantly improved dyslipidemia, particularly serum total cholesterol and LDL cholesterol among HIV-infected patients with dyslipidemia receiving antiretroviral therapy without lipid lowering medication treatment.

Keywords: nutritional counseling, LDL cholesterol, HIV-infected person, Thailand

Introduction

Prior to HAART era, abnormalities in lipid metabolism were demonstrated in HIV-infected patients, including increased triglyceride and decreased total cholesterol, LDL cholesterol and HDL cholesterol levels (Grunfeld, Pang et al. 1992; Feingold, Krauss et al. 1993). An increase in triglyceride and decreased in total cholesterol were associated with advanced HIV disease (Shor-Posner, Basit et al. 1993).

Although intensive antiretroviral therapies dramatically reduce mortality rates in HIV-infected patients (Palella, Delaney et al. 1998), significant metabolic complications associated with antiretroviral therapy treatment such as hyperlipidemia, hyperglycemia and lipodystrophy have been increasingly recognized. The dyslipidemia associated with use of antiretroviral therapy, particularly among individuals receiving HAART that contained a PI, has been commonly reported (Carr, Samaras et al. 1999; Purnell, Zambon et al. 2000; Tsiodras, Mantzoros et al. 2000; Fellay, Boubaker et al. 2001; Dube, Qian et al. 2002). A prospective study in fifty-six HIV-infected patients (Vergis, Paterson et al. 2001) showed that patients adherent to a PI-containing regimen at least 80% of the time were more likely to have increased LDL cholesterol than those who were not adherent (79% vs 26%). Similarly, a cross sectional study in 170 Thai HIV-infected adults (Hiransuthikul, Hiransuthikul et al. 2007) showed that dyslipidemia was prevalent in HIV-infected patients receiving antiretroviral therapy. About half of them (52%) had hypercholesterolemia, 44% had high LDL cholesterol and 43% had hypertriglyceridemia. The study also showed that older aged, male sex and those who received boosted PI were significantly positive associations with total cholesterol and LDL cholesterol.

Dyslipidemia is one of the metabolic complications that have been observed in HIV-infected patients receiving antiretroviral therapy. The prevalence of dyslipidemia in HIV-infected individuals receiving antiretroviral therapy varies from 30% to 80% depending on drug combination and diagnostic definition criteria (Sprinz, Lazzaretti et al. 2010). A cross sectional study in 56 Thai HIV-infected adults showed that dyslipidemia was common in Thai HIV-infected patients receiving antiretroviral therapy. A little more than half of them (53.6%) were observed with dyslipidemia (Chuapai, Kiertiburanakul et al. 2007). It is a well known major risk factor for cardiovascular disease. There is an evidence supported that dyslipidemia in HIV-infected receiving antiretroviral therapy may increase the risk of myocardial infarction. A prospective study in 23, 468 HIV-infected patients (Friis-Moller, Sabin et al. 2003) showed that antiretroviral therapy was associated with an increase of 26% in the rate of myocardial infarction per year of exposure.

The long-term consequences of treatment with lipid-lowering drugs in HIV-infected patients remain unknown. Moreover, there is potential of drug interactions between lipid-lowering medications and antiretroviral drugs. Therefore, therapeutic lifestyle changes (TLC) remain an initial management of dyslipidemia in HIV-infected individuals (NCEP (2002)) (Grundy, Cleeman et al. 2004). Although lipid lowering medications successfully reduced LDL cholesterol and lower risk of coronary heart disease in several large randomized clinical trials (Heart Protection Study (2002); ALLHAT-LLT (2002); Sever, Dahlof et al. 2003). There is no evidence to suggest that HIV-infected patients with hyperlipidemia should be provided more aggressive intervention for lipid management than those intervention used for the general population.

Dietary intervention in HIV-infected persons showed an improvement in important cardiovascular risks in several studies. A prospective study (Sonia Maria de Figueiredo 2013) showed that two sessions of nutritional counseling according to NCEP III, 2001 among 57 HIV-infected patients with antiretroviral therapy. 8.7% reduction of LDL cholesterol during 3 months was observed. A 6-month randomized study in thirty-four HIV-infected patients with metabolic syndrome as defined by NECP (Fitch, Anderson et al. 2006) showed that patients in a weekly individual counseling sessions with a registered dietician group showed a significant reduction in waist circumference (-2.6$\pm$ 1.1 vs 1.2$\pm$ 1.0 cm.) and systolic blood pressure (-13$\pm$ 4 cm. vs 4 $\pm$ 4 cm.), compared to control group. However, the study did not observe any improvement on lipid levels. Similarly, a 12-month randomized clinical trial in fifty-three HIV-infected patients receiving HAART (Luara B. Almeida 2011) did not observe significant changes in lipid profile levels between the nutritional counseling and the control groups, but the study showed that participants in the nutritional counseling group had an improvement in their diet by reducing more fat consumption and increasing fiber intake than the control group. A recent 12-month randomized clinical trial in eighty-three HIV-infected patients, naïve from HARRT at baseline (Lazzaretti, Kuhmmer et al. 2012) showed that diet prevented dyslipidemia associated with antiretroviral therapy among patients received dietary intervention. Of these 83 HIV-infected patients, the intervention group significantly developed hyperlipidemia lesser than the control group (7% vs 39%).

To our knowledge, the studies that have addressed the role of individual nutritional counseling on an improvement dyslipidemia among Thai HIV-infected patients is very

limited. In Thailand, there is currently no nutritional intervention for HIV-infected individuals with dyslipidemia. In addition, most of the existing published-in-English studies were conducted in the western people who consumed different diet patterns from Thai population. Therefore, the present study was conducted in order to determine the effect of nutritional counseling on LDL cholesterol among Thai HIV-infected adults receiving antiretroviral therapy.

Methods

Study design and participants

We conducted a 24-week follow-up, randomized controlled study in HIV-infected patients who were on stable (at least 12 weeks) antiretroviral therapy with dyslipidemia and were eligible to initiate therapeutic lifestyle changes (TLC) according to the National Cholesterol Education Program (NCEP) (2002) (Schambelan, Benson et al. 2002). The aim of the study was to determine whether individual nutritional counseling reduced dyslipidemia, particularly LDL cholesterol, among HIV-infected patients who were on stable ARV with hyperlipidemia above LDL cholesterol goals according to NCEP (2002) guideline. Participants who had severe hyperlipidemia requiring lipid-lowering medication were excluded.

Data for this study was collected between March 2012 and March 2013. Participants were recruited from HIV-infected patients with abnormal LDL cholesterol who had been on antiretroviral therapy at least 3 months and visit outpatient department (OPD) at Bamrasnaradura Hospital. Participants were screened for their lipid profile and risk of

cardiovascular disease (CVD) according to NCEP. Eighty eligible patients were recruited. Of these, eight refused to participate. The reasons for not participating included inconvenience of visiting the hospital frequently or/and being unwilling to come if they did not have appointment with their own physicians. A total of 72 patients were randomized into two groups; 35 patients were in individual counseling with a nutritionist group and 37 patients were in the standard care group. Thirteen withdrew and 59 completed the study at 24 weeks of follow up. Of these 29 (82.9%) and 30 (81.1%) were from intervention and control group, respectively (Fig.1).

Data collection

Prior to conducting interviews and taking measurements, informed consent was obtained from all eligible participants who were willing to participate in the study. At baseline, all participating participants were measured to obtain blood pressure, anthropometric data including weight, height, waist and hip circumference. Participants' dietary intake was assessed with 24-hr recall with trained nutritionists. After that, participants were interviewed about their demographic characteristics, lifestyles behaviors; physical activity and alcohol consumption, and risk of cardiovascular diseases such as family history and smoking with a self-administered questionnaire (see Appendix 2). Self–reported physical activity was assessed using the short interview version of the International Physical Activity Questionnaire (IPAQ) (Booth 2000). Physical activity level was calculated as the product of frequency and the duration of each activity in minutes per week, weighted by an estimate of the metabolic equivalent (MET) of the activity. The result was reported as the mean MET-min per week (Kriska, Knowler et al. 1990).

The participants in the control group received general verbal diet information at baseline and at subsequent annual visit as standard care, which is no individual counseling. Participants were assessed for their dietary intake with 24-hr recall by a nutritionist at baseline and week 24 of the study.

The participants in the intervention group were given detailed advice on reducing saturated fat, trans fat, and cholesterol and increasing soluble fiber, including a Thai manual on nutritional promotion for HIV-infected patients with dyslipidemia (Chotivichien S 2012) and written information of Thai food based dietary guidelines (Bureau of Nutrition, MOPH). There was a total of 7 sessions with a professional nutritionist during the intervention: at baseline, 2 weeks, 4 weeks, 8 weeks, 12 weeks, 18 weeks and 24 weeks. The dietary intervention goals followed the Therapeutic Lifestyle Change (TLC) diet guidelines, which focus on LDL cholesterol changes according to the National Cholesterol Education Program (NCEP) Adult Treatment Panel III (ATP III). Food models were used to assess diet quantity. Participants' dietary intake was assessed with 24-hr recall by a nutritionist in every visit. The dietary advice was provided based on participants' one week food records. Participants were asked to record food they consumed one week prior to the next visit (see Appendix 3). All participants received 200 Baht (~$7) for their participation at each visit. Lipid profile tests were collected and analyzed by technicians at laboratory of Bamrasnaradura hospital. Cost for lipid profile tests at week 12 and 24 of all participants were paid by the study's fund. This study was reviewed and approved by Institutional Review Boards of the University of California at Los Angeles, the Thailand Ministry of

Public Health Ethical Review Committee for Research in Human Subjects, and the Ethics Board of Bamrasnaradura hospital.

Loss to follow up

We asked information about contact phone numbers and addresses of participants. This information was kept confidential. Participants were contacted by phone call to remind them of a follow-up visit. The reminding process was done by one of the research staff about one week before the next follow-up visit. In addition, the staff used the same process to remind any participant who did not come back within a week after his/her appointment. All the processes of reminding and contacting participants were included in the informed consent. Of those 72 participants, 13 (18.1%) were loss to follow up. Among those loss to follow up, 5 (38.5%) could not be reached, 3 (23.1%) moved to other provinces, 3 (23.1%) changed their mind and wanted to withdraw from the study and 2 (15.4%) was busy and did not have time to come to the hospital for the study.

Measurements

Outcome variables

Lipid profiles: changes from baseline (continuous variables)

- **LDL cholesterol**
- **Total cholesterol**
- **HDL cholesterol**
- **Triglyceride**

Participants were asked to fast 12 hours overnight before obtaining the venous blood on the next morning. Lipid profiles (total cholesterol, LDL cholesterol, HDL cholesterol and triglyceride) were measured using enzymatic colorimetric methods at the laboratory of Bamrasnaradura hospital.

Exposures and or covariates

Detailed information regarding these measurements is provided in chapter II (objectives and methods).

Data analyses

Descriptive analysis was used to describe qualitative variables. Each measurement and characteristic of interest at baseline was described as frequency distributions in terms of mean (and standard deviation) in continuous variables or number (percentages) in categorical variables. Differences between groups were compared using the two-sample t-test for continuous variables and Chi-square test or Fisher's exact tests for categorical variables. Nutrient intake, including energy, the amount of saturated fat (SFA), polyunsaturated fat (PUFA), monounsaturated fat was analyzed from dietary intake with the use of the Thai Fatty Acid program, using a Thai computerized dietary database. We analyzed the data in this randomized controlled trial using the 'intention to treat' method. Participants had their LDL cholesterol levels measured at 3 occasions; baseline (week 0), week 12 and week 24. As our primary outcome variable (LDL cholesterol level) changes over time, we used longitudinal methods of analysis that examine these repeated and correlated observation within subject overtime. We used linear mixed models, which

assumes the correlation between repeated measurements arises because each subject has an underlying level of response that persists over time and affects all repeated measurements on that subject (Fitzmaurice Garrett M. 1962-).

$$Yij = X'ij\beta + b_i + \varepsilon ij$$

Where Yij = the vector of responses for the ith subject over the follow-up time points

$X'ij$ = Fixed effects

β = Corresponding regression coefficients of fixed effects

b_i = a random individual-specific effect

εij = a within-individual measurement error

For subject in the standard care group;

$$E(Yij|\ Xij) = \beta_1 + \beta_2 t_j$$

Where β_1 = the mean blood LDL cholesterol level at baseline (week 0)

β_2 = the change in mean blood LDL cholesterol level (mg/dL) per week

Similarly, for subject in the intervention group;

$$E(Yij|\ Xij) = \beta_1 + (\beta_2+\beta_3)t_j$$

Where β_1 = the mean blood LDL cholesterol level at baseline (week 0)

$\beta_2+\beta_3$ = the change in mean blood LDL cholesterol level (mg/dL) per week

Thus, our hypothesis is the two groups (standard care and intervention group) are equally effective in reducing blood LDL cholesterol levels, which is $\beta_3 = 0$

Prior to fitting a model, a graphical summary of this data was plotted in order to illustrate potential covariate effects and sources of variability between-time versus between-subject in the data. We are interested in the differences between the intervention and control group

over time. Thus, we produced separated lines of plots means and standard errors of the mean blood LDL cholesterol level over time. In this data, we have balanced and equally data (lipid profiles were tested 3 times at week 0, week 12 and week 24 on each of 62 subjects). Follow-up visit was made in weeks (0, 12 and 24 weeks) and the outcome is LDL cholesterol (continuous variable). In order to determine the possible variances and correlations for our data, covariance pattern models were fit. After fit various covariance pattern models, compound symmetry (CS) was the best model among other models with the least Akaike Information Criterion (AIC) value. Then we fitted the models for the outcomes of interest using random effects models. Best subset regression was performed for selection the final model. Statistical analyses were performed using the SAS 9.1.3. statistical software package (SAS Institute Inc., Cary, NC).

Results

Of those 72 participants, 62 (86.1%) completed lipid profile testing at baseline, 12 weeks and 24 weeks, 59 (81.9%) completed dietary interview and measurements (blood pressure, bodyweight, waist and hip circumference) at 24 weeks of follow-up. Among 29 participants completing the 24 weeks follow up in the intervention group, 24 (82.8%) completed the 7 sessions protocol. Baseline characteristics of 72 HIV-infected patients, by group are illustrated in table 1. There is no significant difference in baseline characteristics between participants in intervention group and control group. Of 35 participants in intervention group, 65.7% were female. The average age was 42.3 (standard deviation [s.d.] = 6.2). The average bodyweight was 58.0 kilograms (s.d.=9.8). The mean of body mass index (BMI) was 23.0 kg/m^2 (s.d.=3.0). The average waist circumference was 81.2

centimeters (s.d.=8.0). The average hip circumference was 93.0 centimeters (s.d.=5.3). In term of education, 20.0% had less than a high school education, 51.4% had graduated from high school or its equivalent and the remainder (28.6%) had graduated from University or higher. 11.4% had full time job either worked in private company or government. 28.6% worked part time, 37.1% were business owner and 22.9% were unemployed or being as a housewife. 28.5% reported a monthly income equal or less than 5,000 Baht. About half of them (51.4%) reported a monthly income more than 10,000 Baht. Majority of them (80.0%) were ever married. For the lipid profile; the mean of total cholesterol, LDL, HDL and triglyceride were 229.2 mg/dL (s.d.=30.7), 161.0 mg/dL (s.d.=24.2), 56.1 mg/dL (s.d.=18.4) and 152.1 mg/dL (s.d.=89.8%), respectively.

Of 37 participants in control group, approximately half of them (51.4%) were female. The average age was 43.9 (standard deviation [s.d.] = 7.5). The average bodyweight was 62.2 kilograms (s.d.=11.9). The mean of body mass index (BMI) was 23.5 kg/m^2 (s.d.=4.2). The average waist circumference was 84.2 centimeters (s.d.=9.8). The average hip circumference was 94.6 centimeters (s.d.=7.7). The mean of waist hip ratio was 0.89 (s.d.=0.06). In term of education, 18.9% had less than a high school education, 37.8% had graduated from high school or its equivalent and the remainder (43.2%) had graduated from University or higher. 13.5% had full time job either worked in private company or government. 21.6% worked part time, about half of them (51.4%) were business owner and 13.5% were unemployed or being as a housewife. 10.8% reported a monthly income equal or less than 5,000 Baht. More than half of them (59.4%) reported a monthly income more than 10,000 Baht. Most of them (64.9%) were ever married. For the lipid profile; the mean

of total cholesterol, LDL, HDL and triglyceride were 226.4 mg/dL (s.d.=21.8), 157.3 mg/dL (s.d.=21.6), 54.2 mg/dL (s.d.=17.2) and 166.2 mg/dL (s.d.=99.1%), respectively.

For dietary intake parameters, the percentage of total calories obtained from fat was about 27% in both groups. No significant differences between groups were seen in term of dietary parameters (including energy, %protein, %carbohydrate, %fat, %SFA, %PUFA,%MUFA, total fiber and soluble fiber). The average daily total fiber intake was 7.23 g (s.d.=3.78) and 6.28 (s.d.=4.46) in the intervention group and the control group, respectively. For physical activity, there is no significant difference in baseline average total physical activity between both groups measured in term of met-min/week (Table 2). Overall, both groups showed downward trend of mean of LDL cholesterol at the 24 weeks of follow up compared to baseline. The intervention group showed deeper decline in slope than the control group. After 12 weeks of follow up, a slightly decreasing trend is continuously observed from week 12 to week 24 in the intervention group, whereas there is no obvious change in mean LDL cholesterol is discerned in the control group (Figure 2). These results as well as those in tables 3 to 6 are based on the linear mixed models described in the methods section.

Table 3, the percentage of total calories obtained from fat was approximately 27% among 59 participants who completed dietary interview at 24 weeks. Of total calories, about 5%, 2% and 4% was obtained from SFA, PUFA and MUFA, respectively. Participants in the intervention group demonstrated a significant reduction in energy intake (-244.1 $\pm$100.2 kcal/d, p=0.02) and in carbohydrate intake (-38.3 $\pm$ 16.7 g/day, p=0.03). While a greater

reduction in mean fat intake was demonstrated in the intervention group than the control group (19% vs 4%) at 24 weeks, no significant differences between these two groups were observed in mean reduction of fat intake. We observed a significant increase in total fiber intake in the control group (3.2 $\pm$ 1.1 g/day, p=0.004) but no significant changes were observed in energy intake (-28.3 $\pm$ 98.5 kcal/d, p=0.78) and other diet parameters. Compared the difference of changes with the control group at 24 weeks of follow up, no significant changes were seen between these two groups in any diet parameters (Table 4).

Table 5, compared between baseline and 24 weeks of follow up, participants in the intervention group demonstrated a significant reduction in bodyweight (-1.2 $\pm$ 0.4 kgs, p=0.007). Both intervention and control group demonstrated a significant reduction in hip circumference (-1.7 $\pm$ 0.5 cm, p=0.002 versus -1.1 $\pm$ 0.5 cm, p=0.02, respectively). A slight increase in waist hip ratio was observed in the intervention group from 0.88 to 0.89 (p=0.01). There was no overall change in blood pressure in either group. Comparing the difference of changes between the intervention and the control group at 24 weeks of follow up, no significant changes were seen between these two groups in any anthropometric and blood pressure parameters.

Compared with the control group, participants in the intervention group demonstrated a significant reduction in serum total cholesterol (-19.0 $\pm$ 4.6 versus 0.2 $\pm$4.3 mg/dL, p= <.0001) and LDL cholesterol (-21.5 $\pm$ 4.1 versus -6.8 $\pm$ 3.8 mg/dL, p= <.0001) at 24 weeks of follow up. The difference of changes between these two groups at 24 weeks was also demonstrated in total cholesterol (19.2 $\pm$ 6.3 mg/dL , p=0.003) and LDL cholesterol (14.7 $\pm$

5.5 mg/dL, p=0.009). There were no significant changes in HDL cholesterol and triglyceride level. At 12 weeks of follow up, participants in the intervention group demonstrated a significant reduction in total cholesterol (-14.4 + 4.6 mg/dL, p=0.002), in LDL cholesterol (-13.7 + 4.1 mg/dL, p=0.001), and in triglyceride (-30.4 + 13.8 mg/dL, p=0.03) whereas participants in the control group showed significant reduction only in LDL cholesterol (-7.7 + 3.8 mg/dL, p=0.04). However, there were no significant differences of the changes of mean lipid levels between the intervention and the control groups at 12 weeks of follow up (Table 6).

Table 7 demonstrated the number and percentage of participants who adhered to dietary recommendations according to Therapeutic Lifestyle changes (TLC) of the National Cholesterol Education Program (NCEP) (2002). We adapted the NCEP guideline for percent fat to make it appropriate for Thais. The NCEP recommends 25-35% of total calories for total fat. But in this present study, the recommendation for total fat we suggested were less than 25%. The rationale for reducing percentage of total fat was that dietary patterns for Thais contain less fat than for Western population. Compared to baseline, a fewer percentage of participants in the intervention group than the control group reported adherence to the dietary recommendation at 24 weeks regarding percent fat, SFA, PUFA and MUFA were observed. Almost none of participants in either group consumed 20-30 gram a day of total fiber. Compared to the control group, participants in the intervention group demonstrated a greater adherence in appropriate consumption of protein and carbohydrate at 24 weeks of follow up.

Table 8 demonstrated body mass index (BMI) of participants who had weight loss at least 1 kilogram at 24 weeks of follow up. Compared to baseline, mean BMI of both groups were in a normal range (18.5-24.9 kg/m^2) for baseline and at 24 weeks. Of 14 participants who had weight loss in each group, 72% of participants in the intervention group was in a normal range of BMI at baseline and increased to 86% at 24 weeks of follow up, whereas half of participants (50%) in the control group was in a normal range of BMI at baseline but decreased to 43% at 24 weeks of follow up. In the intervention group, the weight loss was from obese to preobese and preobese to normal. No increase in underweight. In the control group, the weight loss was from obese to preobese and normal to underweight.

Discussion

This study is one of the few to assess individual nutrition counseling in Thai HIV-infected persons. A small non-randomized study (n=13) showed individual counseling with a nutritionist improved nutritional status of Thai HIV-infected patients after 5 sessions in 12 weeks (Krongthaew 2003). The present study is the first that explored effect of nutritional counseling and metabolic complication parameters, particularly low density lipoprotein cholesterol (LDL-C) in Thailand.

Our study found a significant difference in mean reduction from baseline of total cholesterol and LDL-C between the intervention group (receiving individual counseling from a nutritionist) and the control group (receiving usual care). Compared to the control group, patients in the intervention showed significant improvement in serum total cholesterol (8% vs 0%) and LDL-C (13% vs 4%) at 24 weeks of follow up. Only the

intervention group showed a reduction in triglyceride at 24 weeks (11%) whereas the control group showed a 6% increase in triglyceride from baseline. At 12 weeks of follow up, a reduction in total cholesterol (6% vs 3%), LDL-C (8% vs 5%) and triglyceride (18% vs 7%) were observed in the intervention and the control group, respectively. However, none of these differences between two groups at 12 weeks of follow up were significant. Our findings are consistent with a three months non-randomized study in 57 HIV-infected patients (Sonia Maria de Figueiredo 2013) that nutritional counseling based on the NCEP guideline improved dyslipidemia induced by antiretroviral therapy in HIV-infected individuals by significantly lowering in lipid levels, particularly total cholesterol and LDL-C levels. Likewise, no significant difference was observed in total fiber, PUFA and MUFA after the dietary intervention.

Our findings are consistent with results of multiple randomized controlled trials in demonstrating that counseling with a nutritionist effectively improved lipid levels in non HIV-infected patients in Western countries (Ramsay, Yeo et al. 1991; Hunninghake, Stein et al. 1993; Geil, Anderson et al. 1995; Rhodes, Bookstein et al. 1996). However, our study found lower reduction in total cholesterol and LDL-C compared to Rhodes et al. study. The author (Rhodes, Bookstein et al. 1996) reported a reduction in total cholesterol (10% vs 7%) and LDL-C (11% vs 9%) at 12 weeks of follow up between dietitian and non-dietitian groups, respectively. But we found greater reduction in total cholesterol than the level reported in some study (Ramsay, Yeo et al. 1991), where the average reduction in serum cholesterol concentration ranged from 0% to 4% over six months to six years. The average reduction in lipid levels from our study was similar to a 9-week randomized controlled trial

in 111 HIV-naïve patients (Hunninghake, Stein et al. 1993), with a reduction of 5% in total cholesterol level in nutritional counseling group.

Furthermore, our study is consistent with other studies that counseling effectively improved lipid levels and other cardiovascular risk factors in patients not receiving lipid-lowering drugs (Ockene, Hebert et al. 1999; Henkin, Shai et al. 2000; Yorioka, Masaki et al. 2000; Barrios, Blanco et al. 2002). Despite our finding of a significant improvement in lipid levels in the intervention group, we did not observe significant difference in fat intake (%fat, %SFA, %PUFA and %MUFA) between groups. This may be explained by the fact that both groups in our study were consuming low percentage of saturated fat and cholesterol content at the baseline in comparison with Western populations. Fiber intake was significantly increased among participants in the control group (3.2 $\pm$ 1.1 g/day, p=0.004). At 24 weeks of follow up, a reduction in fat intake (19% vs 4%) and cholesterol consumption (10% vs 10%) were observed in the intervention and the control group, respectively. A significant decrease in total cholesterol and LDL-C in this study might be the result of a decline in total energy consumption (-224.1 $\pm$ 100.2 kcal/day, p=0.02) and a significant reduction in weight (-1.2 $\pm$ 0.4 kgs, p=0.007). Participants in the intervention group decreased their intake of carbohydrate at 24 weeks of follow up (-38.3 $\pm$ 16.7 g/day, p=0.03). In contrast to our study, a 12-month randomized controlled trial of 53 HIV-infected patients receiving antiretroviral therapy in Brazil showed a reduction in fat consumption at the end of study but no significant differences in lipid levels between groups. Similarly to our findings, this study (Luara B. Almeida 2011) demonstrated no significant changes in waist circumference and blood pressure between groups.

Weight loss and energy reduction amongst individual that are undernourished might have detrimental effects in HIV-infected patients if they did not consume adequate energy intake to maintain their desirable body weight. Energy requirement in HIV-infected adults has been suggested to increase by 10% to maintain bodyweight and physical activity (WHO, 2003). Unfortunately, we did not have data on HIV viral load and CD4+ of participants at 24 weeks of follow up to assess their health with regard to HIV infection. However, we did not observe any health deterioration among participants in our study. None of the participants who reported weight loss reported illness or were admitted to the hospital during the time of follow up. Baseline data of these participants showed that they were asymptomatic HIV-infected patients and had high CD4+ levels (average CD4+ of 496 cells/mm^3 as they have been treated with antiretroviral therapy for at least 3 months). We could not identify a negative impact of the reported weight loss and caloric reduction in the intervention group. We suspect that individual nutritional counseling resulted in a favorable reduction in weight that improved nutritional status of participants. The weight loss was from BMI of obese to preobese, from preobese to normal range and no increase in underweight at 24 weeks of follow up. Nevertheless, adherences to dietary change might be temporary and tend to reduce as time passes. A 2-year randomized controlled trial (Greenberg, Stampfer et al. 2009) showed that dietary adherence declined from 81% at the first month of dietary intervention to 57% at 24 months. Therefore, a strategy that helps people maintain dietary intake as recommended should be considered.

Our study did not find protective effects in term of reduction in waist circumference and blood pressure, which contrasts with other studies (Moyle, Lloyd et al. 2001; Kathleen V.

Fitch 2006). It is important to note that participants in those studies presented with high body mass index (BMI) and waist circumferences at baseline whereas participants in our study presented with normal BMI and waist circumference at baseline. We found a slight increase in waist-hip ratio for both groups; this finding is similar to some previous study (Lazzaretti, Kuhmmer et al. 2012). In our study, hip circumference was significantly reduced in both groups at 24 weeks. This finding may be explained by lipodystrophy (fat redistribution) in HIV-infected individuals receiving antiretroviral therapy. Lipodystrophy may present with body changes such as localized accumulation (lipohypertrophy) and/or with loss (lipoatrophy) in body fat (WHO 2004).

The strengths of our study include the use of a randomized controlled trial (RCT), with a follow up period long enough to observe the changes in lipid levels. The RCT not only avoids the reverse causation compared to case control or cross-sectional designs but also reduces the risk of a serious imbalance in known and unknown factors that could influence the clinical course of the patients. Furthermore, our study assessed the effect of nutritional counseling in metabolic changes among HIV-infected individuals without lipid lowering medication since there is little evidence of nutritional intervention therapy for managing metabolic abnormalities in HIV-infected patients on antiretroviral therapy in Thailand. This study demonstrated a practical nutritional counseling program for Thai settings that proved to be effective in improving dyslipidemia in HIV-infected patients.

Some limitations in this study should be noted. The study has a relatively small sample size that may result in low statistical power to detect a small difference such as in diet

parameters. Our study found no significant differences of the changes of mean or percentage of dietary consumption between groups at 24 weeks of follow up. There is a possibility of contamination of the information i.e., the control group may receive information about lifestyle modification from talking with health personnel or learn from other sources such as the Internet. Neither group reported a significant increase in physical activity during the follow up period. Overall physical activity from self-report showed no significant differences between two groups. Thus, the findings of reducing lipid profile levels, body weight and increase fiber intake in the intervention group from this study is less likely due to the possibility that participants in the intervention group increased their physical activity during follow up period. It has been reported that soluble fiber significantly lowered total cholesterol and LDL-C (Brown, Rosner et al. 1999) ((NHLBI), 2006). In this study, we could not evaluate the effect of soluble fiber and trans fat because the Thai food database that we used to analyze this data did not contain these parameters. The values of soluble fiber presented in this study were based on values of some Thai food items that were analyzed by Bureau of Nutrition, Thailand (Bureau of Nutrition, 2012). Lastly, we used a single 24-hour recall to assess dietary intakes of participants because it is practical and easily collected (Barrett-Connor 1991). However, this method results in inflated variance and may not be sensitive enough to detect the effects of our dietary intervention.

In conclusion, this study underscored the benefit of intensive dietary intervention, sessions of individual counseling with a nutritionist, on dyslipidemia improvement among HIV-infected patients receiving antiretroviral therapy, without lipid medication treatment and for

the first time demonstrated this benefit in a Thai population. The effect we observed from our study indicated a significant reduction in LDL-C level attributed to individual counseling during 24 weeks follow-up. The effect of lowering LDL-C in our study may be explained by the weight reduction and a decrease in total energy consumption. Although the effect of lowering in LDL-C in our study was small, we found greater differences in mean LDL-C between the intervention group and the control group when time passed and a greater reduction in LDL-C level was observed in the intervention group. A longer follow up period may demonstrate a clearer effect of nutritional intervention on the lipid profile. Our study population was low (limited by difficulty in recruitment and widespread use of lipid-lowering medication) and a larger population may be required to demonstrate changes in some parameters.

The findings from our study may have practical implications for clinicians in planning a better care and treatment for HIV-infected patients. To date, long-term consequences of treatment with lipid-lowering drugs in HIV-infected patients are unknown. To avoid potential of drug interactions between lipid-lowering medications and antiretroviral drugs, lifestyle modification is recommended as an initial management of dyslipidemia in HIV-infected individuals. Our study demonstrated a greater reduction in LDL-C levels in patients who received diet counseling from a nutritionist. Thus, this proposed intervention reinforces the benefit of individual nutrition counseling for HIV-infected patients with dyslipidemia and should be integrated as part of care in HIV clinic.

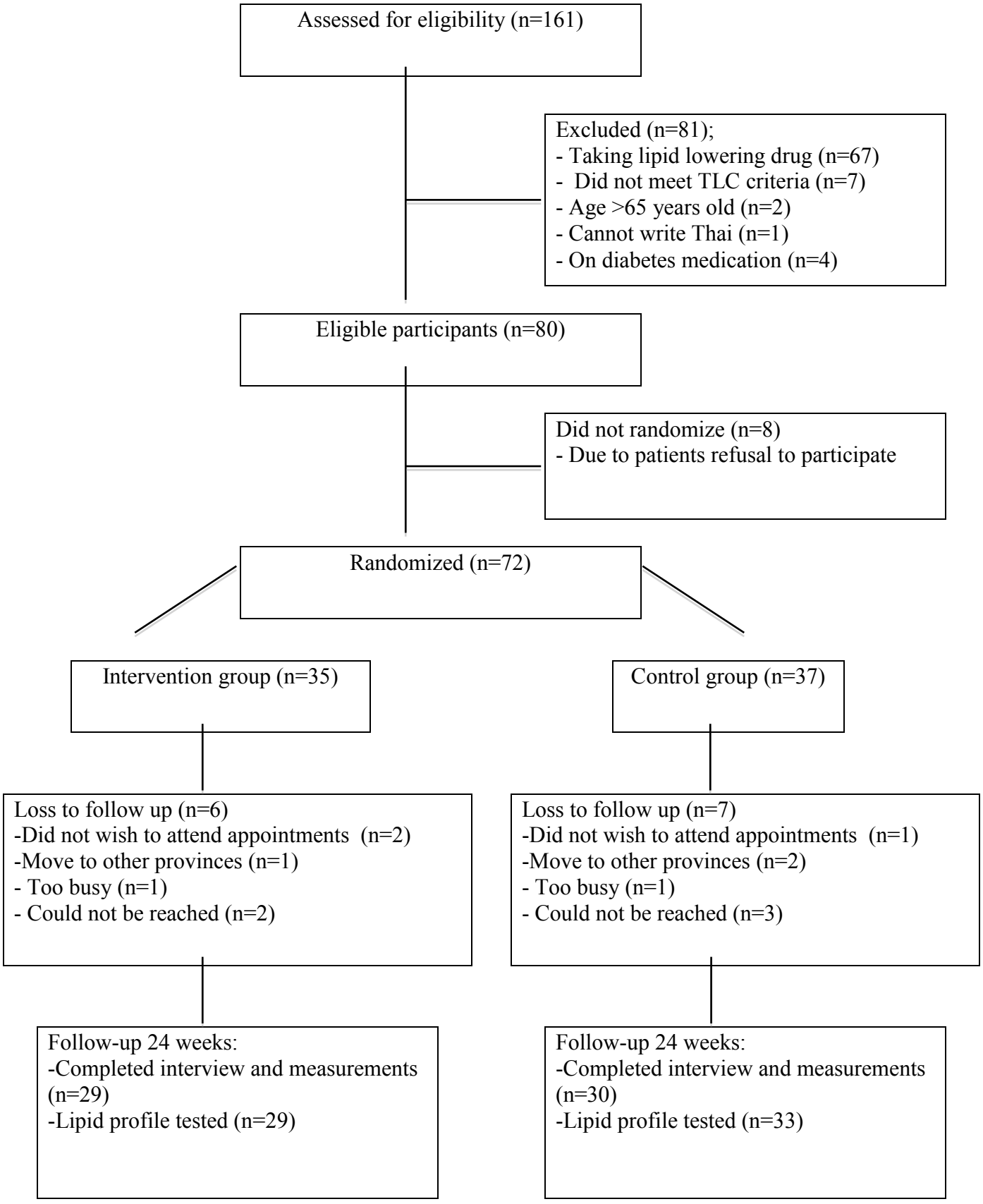

Figure 1. Diagram demonstrating flow of participants through the study

Table 1. Baseline characteristics of participants (N=72)

Characteristics	Total (N=72) Mean (s.d.*); N (%)	Intervention group (N=35) Mean (s.d.*); N (%)	Control group (N=37) Mean (s.d.*); N (%)	p-value
Age (years)	43.1 (6.9)	42.3 (6.2)	43.9 (7.5)	0.263
Female gender – N (%)	42 (58.3%)	23 (65.7%)	19 (51.4%)	0.217
Anthropometric measurement				
Bodyweight (kgs)	60.2 (11.0)	58.0 (9.8)	62.2 (11.9)	0.114
Body Mass Index (kg/m^2)	23.3 (3.7)	23.0 (3.0)	23.5 (4.2)	0.517
Waist circumference (cm)	82.7 (9.0)	81.2 (8.0)	84.2 (9.8)	0.161
Hip circumference (cm)	93.9 (6.7)	93.0 (5.3)	94.6 (7.7)	0.306
Waist hip ratio	0.88 (0.06)	0.87 (0.05)	0.89 (0.06)	0.24
Education – N (%)				0.678
Less than a high school	14 (19.4%)	7 (20.0%)	7 (18.9%)	
High school or its equivalent	32 (44.4%)	18 (51.4%)	14 (37.8%)	
University or higher	26 (36.1%)	10 (28.6%)	16 (43.2%)	
Occupation – N (%)				0.754
Full time: Government or Private	9 (12.5%)	4 (11.4%)	5 (13.5%)	
Part time: Regular or irregular	18 (25.0%)	10 (28.6%)	8 (21.6%)	
Owner of business	32 (44.4%)	13 (37.1%)	19 (51.4%)	
Unemployed	13 (18.1%)	8 (22.9%)	5 (13.5%)	
Married/ever married – N (%)	52 (72.2%)	28 (80.0%)	24 (64.9%)	0.365

*s.d. = Standard Deviation

Table 1. (cont.) Baseline characteristics of participants (N=72)

Characteristics	Total (N=72) Mean (s.d.*); N (%)	Intervention group (N=35) Mean (s.d.*); N (%)	Control group (N=37) Mean (s.d.*); N (%)	p-value
Income (Baht) – N (%)				0.328
5,000 or less	14 (19.4%)	10 (28.6%)	4 (10.8%)	
>5,000 – 10,000	18 (25.0%)	7 (20.0%)	11 (29.7%)	
>10,000	40 (55.6%)	18 (51.4%)	22 (59.5%)	
Smoke cigarette – N (%)	9 (12.5%)	4 (11.4%)	5 (13.5%)	1.000
Drink alcohol – N (%)	21 (29.2%)	11 (31.4%)	10 (27.0%)	0.681
Treatment with hypertension – N (%)	6 (8.3%)	3 (8.6%)	3 (8.1%)	1.000
Duration of being known as HIV positive (years)	9.6 (4.5)	10.1 (5.0)	9.2 (3.9)	0.399
Route of HIV acquisition – N (%)				0.712
Heterosexual sex	58 (80.6%)	28 (80.0%)	30 (81.1%)	
Homosexual sex	9 (12.5%)	5 (14.3%)	4 (10.8%)	
Intravenous drug	1 (1.4%)	1 (2.9%)	0	
Bisexual sex	2 (2.8%)	0	2 (2.8%)	
Tattoo	2 (2.8%)	1 (2.9%)	1 (2.7%)	
CD4 count (cells/mm^3)	496.0 (177.7)	517.8 (183.6)	475.4 (171.9)	0.315
Current ARV treatment – N (%)				
Treatment with protease	12 (16.7%)	5 (14.3%)	7 (18.9%)	0.598
Treatment with NRT	70 (97.2%)	34 (97.1%)	36 (97.3%)	1.000
Treatment with NNRT	62 (86.1%)	30 (85.7%)	32 (86.5%)	1.000

*s.d. = Standard Deviation

Table 2. Baseline lipid profile, dietary intake and physical activity of participants in the study

Characteristics	Total (N=72) Mean (s.d.*)	Intervention group (N=35) Mean (s.d.*)	Control group (N=37) Mean (s.d.*)	p-value
Lipid profile				
Total Cholesterol (mg/dL)	227.8 (26.4)	229.2 (30.7)	226.4 (21.8)	0.653
LDL-C (mg/dL)	159.1 (22.8)	161.0 (24.2)	157.3 (21.6)	0.502
HDL-C (mg/dL)	55.2 (17.7)	56.1 (18.4)	54.2 (17.2)	0.652
Triglyceride (mg/dL)	159.3 (94.3)	152.1 (89.8)	166.2 (99.1)	0.531
Dietary parameters				
Energy (kcal)	1364.8 (487.0)	1362.9 (543.3)	1366.7 (434.8)	0.974
Protein (%)	14.7 (4.6)	14.1 (5.1)	15.2 (4.0)	0.322
Carbohydrate (%)	57.7 (10.1)	58.6 (11.3)	56.9 (8.9)	0.478
Fat (%)	27.7 (8.8)	27.3 (10.3)	28.0 (7.3)	0.763
Saturated fat (%)	4.9 (4.5)	4.8 (3.9)	4.9 (5.1)	0.929
Polyunsaturated fat (%)	2.0 (2.0)	2.5 (2.2)	1.6 (1.7)	0.072
Monounsaturated fat (%)	4.4 (3.8)	4.6 (3.9)	4.3 (3.8)	0.741
Cholesterol (mg)	187.2 (149.2)	173.0 (144.8)	200.6 (154.0)	0.437
Total fiber (g)	6.7 (4.1)	7.2 (3.8)	6.3 (4.5)	0.332
Soluble fiber (mg)	0.4 (0.8)	0.5 (0.9)	0.3 (0.7)	0.281
Physical Activity parameters				
Total PA (Met-min/week)**	448.3 (431.2)	491.9 (477.2)	412.9 (393.0)	0.460

*s.d. = Standard Deviation

**Metabolic equivalent: Met-min calculated by multiply minutes of the respective activity in the past week by 8 (vigorous), 4 (moderate), and 3.3 (walking). Total PA (physical activity) is the sum of vigorous, moderate and walking Met-min.

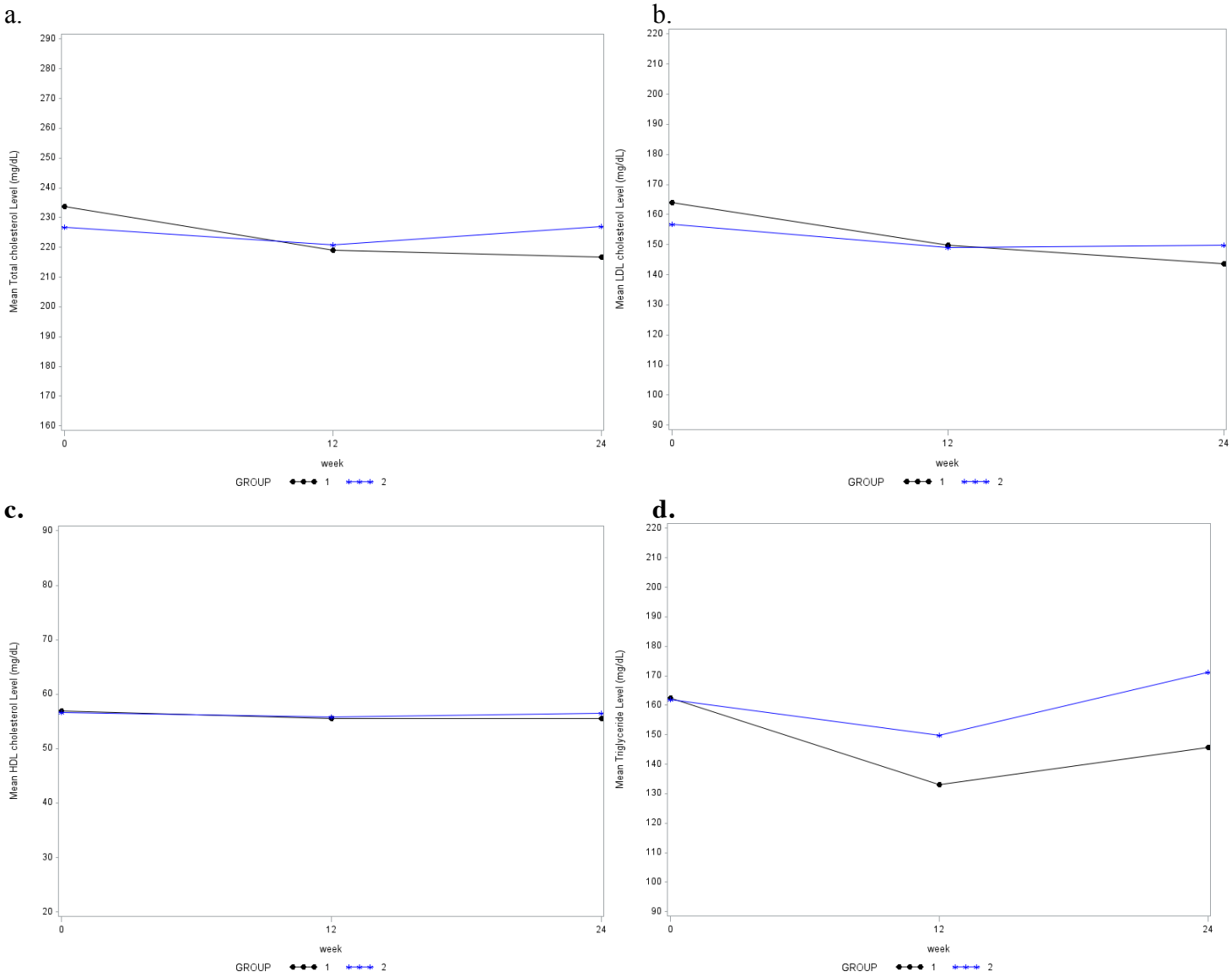

Figure 2. Mean lipid profiles compares between intervention (group 1) and control group (group 2) during 24 weeks of follow-up (mean total cholesterol, LDL-C, HDL-C and triglyceride levels are illustrated in a, b, c and d, respectively.)

Table 3. Dietary intakes between baseline and at 24 weeks of follow up in the intervention and the control groups who completed dietary interview and measurement at 24 weeks

Nutrient intake	Intervention group (N=29) Mean (S.E*)		Control group (N=30) Mean (S.E*)	
	Baseline	Week 24	Baseline	Week 24
Energy (kcal)	1393.8 (84.7)	1149.7 (84.7)	1314.7 (83.3)	1286.4 (83.3)
Protein (g)	47.4 (3.5)	43.6 (3.5)	49.7 (3.5)	45.7 (3.5)
(%) of total energy	14.1 (0.8)	15.3 (0.8)	15.2 (0.8)	14.4 (0.8)
Fat (g)	42.6 (3.6)	34.3 (3.6)	39.3 (3.6)	37.9 (3.6)
(%) of total energy	26.8 (1.5)	26.7 (1.5)	27.3 (1.5)	25.9 (1.5)
Carbohydrate (g)	205.2 (13.5)	166.9 (13.5)	191.2 (13.3)	191.1 (13.3)
(%) of total energy	59.1 (1.7)	58.0 (1.7)	57.5 (1.7)	59.7 (1.7)
SFA (g)	7.7 (1.3)	7.5 (1.3)	7.1 (1.3)	6.5 (1.3)
(%) of total energy	4.7 (0.8)	5.9 (0.8)	5.1 (0.7)	4.4 (0.7)
PUFA (g)	3.7 (0.8)	4.3 (0.8)	2.3 (0.8)	4.1 (0.8)
(%) of total energy	2.3 (0.5)	3.6 (0.5)	1.6 (0.5)	2.7 (0.5)
MUFA (g)	7.5 (1.1)	7.0 (1.1)	6.0 (1.1)	6.7 (1.1)
(%) of total energy	4.6 (0.7)	5.6 (0.7)	4.3 (0.6)	4.5 (0.6)
Cholesterol (mg)	167.3 (25.1)	150.0 (25.1)	192.2 (24.7)	172.6 (24.7)
Total fiber (g)	7.8 (0.8)	9.3 (0.8)	5.7 (0.8)	8.9 (0.8)
Soluble fiber (mg)	0.5 (0.1)	0.3 (0.1)	0.3 (0.1)	0.3 (0.1)

*S.E.=Standard error

Table 4. Differences in mean (S.E.) change scores between the intervention and control groups in dietary intake parameters during baseline and 24 weeks of follow-up

Variable	Difference in mean (S.E.) between week 24 and baseline				Change between two groups at 24 week	p value
	Intervention group (n=29)	p value	Control group (n=30)	p value		
Energy (kcal)	-244.1 (100.2)	**0.018**	-28.3 (98.5)	0.775	215.8 (140.5)	0.129
Protein (g)	-3.7 (3.7)	0.318	-4.1 (3.6)	0.269	-0.3 (5.2)	0.95
Protein (%)	1.1 (0.9)	0.206	-0.8 (0.9)	0.362	-1.9 (1.2)	0.125
Fat (g)	-8.2 (4.7)	0.085	-1.4 (4.6)	0.756	6.8 (6.6)	0.307
Fat (%)	-0.09 (2.2)	0.968	-1.4 (2.1)	0.525	-1.3 (3.1)	0.676
Carbohydrate (g)	-38.3 (16.7)	**0.026**	-0.04 (16.4)	0.998	38.3 (23.5)	0.108
Carbohydrate (%)	-1.1 (2.4)	0.661	2.2 (2.3)	0.357	3.2 (3.3)	0.338
SFA (g)	-0.3 (1.6)	0.879	-0.7 (1.6)	0.689	-0.4 (2.3)	0.863
SFA (%)	1.3 (1.0)	0.214	-0.7 (1.0)	0.464	-2.0 (1.4)	0.163
PUFA (g)	0.6 (1.1)	0.580	1.8 (1.1)	0.111	1.2 (1.6)	0.463
PUFA (%)	1.3 (0.8)	0.090	1.1 (0.7)	0.128	-0.2 (1.0)	0.884
MUFA (g)	-0.4 (1.5)	0.781	0.7 (1.5)	0.640	1.1 (2.1)	0.599
MUFA (%)	1.1 (0.9)	0.246	0.1 (0.9)	0.876	-1.0 (1.3)	0.471
Cholesterol (mg)	-17.3 (34.5)	0.619	-19.6 (34.0)	0.566	-2.3 (48.4)	0.962
Total fiber (g)	1.5 (1.1)	0.175	3.2 (1.1)	**0.004**	1.7 (1.5)	0.270
Soluble fiber (mg)	-0.2 (0.2)	0.316	-0.04 (0.2)	0.797	0.1 (0.2)	0.574

Table 5. Differences in mean (S.E.) change scores between the intervention and control group in anthropometric and blood pressure parameters during baseline and 24 weeks of follow-up

Variable	Intervention group (n=29)		p value	Control group (n=30)		p value	Change between two groups at 24 week (S.E.)	p value
	Mean (S.E.)	Difference from baseline		Mean (S.E.)	Difference from baseline			
Bodyweight (kg)								
Week 0	58.4 (2.3)			60.9 (2.0)				
Week 24	57.2 (2.3)	-1.2 $\pm$ 0.4	**0.007**	60.3 (2.0)	-0.5 $\pm$ 0.4	0.156	0.7 $\pm$ 0.6	0.254
BMI (kg/m^2)								
Week 0	23.2 (0.8)			23.0 (0.7)				
Week 24	22.7 (0.8)	-0.5 $\pm$ 0.2	**0.005**	22.8 (0.7)	-0.2 $\pm$ 0.2	0.120	0.3 $\pm$ 0.2	0.265
Waist (cm)								
Week 0	81.8 (1.9)			82.9 (1.7)				
Week 24	81.8 (1.9)	0.01 $\pm$ 0.7	0.991	82.5 (1.7)	-0.4 $\pm$ 0.6	0.556	-0.4 $\pm$ 1.0	0.688
Hip (cm)								
Week 0	93.2 (1.4)			94.1 (1.3)				
Week 24	91.5 (1.4)	-1.7 $\pm$ 0.5	**0.002**	93.0 (1.3)	-1.1 $\pm$ 0.5	**0.019**	0.5 $\pm$ 0.7	0.444
Waist Hip Ratio								
Week 0	0.88 (0.01)			0.88 (0.01)				
Week 24	0.89 (0.01)	0.02 $\pm$ 0.01	**0.011**	0.89 (0.01)	0.01 $\pm$ 0.01	0.170	-0.01 $\pm$ 0.01	0.306

Table 5. (cont.) Differences in mean (S.E.) change scores between the intervention and control group in anthropometric and blood pressure parameters during baseline and 24 weeks of follow-up

Variable	Intervention group (n=29)		p value	Control group (n=30)		p value	Change between two groups at 24 week (S.E.)	p value
	Mean (S.E.)	Difference from baseline		Mean (S.E.)	Difference from baseline			
Systolic BP (mmHg)								
Week 0	119.2 (2.7)			130.8 (2.7)				
Week 24	121.4 (2.7)	2.2 ± 2.2	0.306	129.3 (2.7)	-1.5 ± 2.1	0.485	-3.7 ± 3.1	0.224
Diastolic BP (mmHg)								
Week 0	75.1 (2.1)			82.5 (2.0)				
Week 24	76.5 (2.1)	1.3 ± 1.7	0.432	82.9 (2.0)	0.5 ±1.6	0.775	-0.8 (2.3)	0.718

Table 6. Differences in mean (S.E.) change scores between the intervention and control group in lipid profiles during baseline, 12 weeks and 24 weeks of follow-up

Variable	Intervention group (n=29)		p value	Control group (n=33)		p value	Change between the two groups (S.E.)	p value
	Mean (S.E.)	Difference from baseline		Mean (S.E.)	Difference from baseline			
Total cholesterol								
Week 0	234.6 (5.5)			226.8 (5.1)				
Week 12	220.3 (5.5)	-14.4 (4.6)	**0.002**	220.8 (5.1)	-6.0 (4.3)	0.165	8.4 (6.3)	0.186
Week 24	215.7 (5.5)	-19.0 (4.6)	**<.0001**	227.0 (5.1)	0.2 (4.3)	0.960	19.2 (6.3)	**0.003**
LDL-C								
Week 0	164.8 (4.9)			156.7 (4.6)				
Week 12	151.1 (4.9)	-13.7 (4.1)	**0.001**	149.0 (4.6)	-7.7 (3.8)	**0.042**	6.0 (5.5)	0.285
Week 24	143.3 (4.9)	-21.5 (4.1)	**<. 0001**	149.9 (4.6)	-6.8 (3.8)	0.072	14.7 (5.5)	**0.009**
HDL-C								
Week 0	56.5 (3.3)			56.6 (3.0)				
Week 12	55.4 (3.3)	-1.1 (1.6)	0.475	55.8 (3.0)	-0.8 (1.5)	0.592	0.4 (2.2)	0.870
Week 24	54.4 (3.3)	-2.1 (1.6)	0.189	56.6 (3.0)	-0.03 (1.5)	0.984	2.1 (2.2)	0.340
Triglyceride								
Week 0	165.5 (17.3)			161.8 (15.9)				
Week 12	135.2 (17.3)	-30.4 (13.8)	**0.030**	149.9 (15.9)	-11.9 (12.7)	0.352	18.5 (18.8)	0.326
Week 24	148.0 (17.3)	-17.5 (13.8)	0.206	171.3 (15.9)	9.5 (12.7)	0.457	27.0 (18.8)	0.152

Table 7. Number and percentage of participants adhering to each dietary recommendation

Recommended Dietary Intake*	Intervention (N=29); n (%)		Control (N=30); n (%)	
	Week 0	Week 24	Week 0	Week 24
1. Reduce percent fat to less than 25% of total calories	13 (44.8%)	14 (48.3%)	13 (43.3%)	17 (56.7%)
2. Reduce Cholesterol to < 200 mg a day	19 (65.5%)	20 (69.0%)	17 (56.7%)	21 (70.0%)
3. Reduce SFA to <7% of total calories	24 (82.8%)	17 (58.6%)	23 (76.7%)	22 (73.3%)
4. Increase PUFA up to 10% of total calories[#]	NA	12 (41.4%)	NA	16 (53.3%)
5. Increase MUFA up to 20% of total calorie[##]	NA	14 (48.3%)	NA	16 (53.3%)
6. Increase total fiber up to 20-30 gram a day	0	1 (3.4%)	0	1 (3.3%)
7. Consume protein approximately 15% of total calories**	2 (6.9%)	8 (27.6%)	4 (13.3%)	5 (16.7%)
8. Consume carbohydrate 50-60% of total calories	10 (34.5%)	10 (34.5%)	8 (26.7%)	7 (23.3%)

*Adapted from the NCEP ATP III guideline (2002)

[#] Increase PUFA consumption and PUFA less than 10% of total calories at 24 weeks of follow up ($(PUFA_{week24} - PUFA_{week0} > 0)$ and PUFA at week 24 $\leq$ 10%)

[##] Increase MUFA consumption and MUFA less than 20% of total calories at 24 weeks of follow up ($(MUFA_{week24} - MUFA_{week0} > 0)$ and MUFA at week 24 $\leq$ 20%)

**Consume protein between 14.5% and 16.0% of total calories (%Protein > 14.5% and < 16%)

Table 8. Body mass index (BMI) of participants who lost weight at least 1 kg at 24 weeks of follow up

Variables	Intervention (N=14); Means (s.d), n (%)		Control (N=14); Means (s.d), n (%)	
	Week 0	**Week 24**	**Week 0**	**Week 24**
BMI (kg/m^2)	23.2 (3.1)	22.1 (2.7)	24.2 (5.3)	23.3 (5.2)
BMI (kg/m^2) – N(%)				
<18.5 [underweight]	1 (7.1%)	1 (7.1%)	2 (14.3%)	3 (21.4%)
18.5-24.9 [normal range]	10 (71.4%)	12 (85.7%)	7 (50.0%)	6 (42.9%)
25-29.9 [preobese]	2 (14.3%)	1 (7.1%)	2 (14.3%)	3 (21.4%)
≥ 30 [Obesity]	1 (7.1%)	0	3 (21.4%)	2 (14.3%)

References

(2002). "Major outcomes in moderately hypercholesterolemic, hypertensive patients randomized to pravastatin vs usual care: The Antihypertensive and Lipid-Lowering Treatment to Prevent Heart Attack Trial (ALLHAT-LLT)." JAMA : the journal of the American Medical Association 288(23): 2998-3007.

(2002). "MRC/BHF Heart Protection Study of cholesterol lowering with simvastatin in 20,536 high-risk individuals: a randomised placebo-controlled trial." Lancet 360(9326): 7-22.

(2002). "Third Report of the National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel III) final report." Circulation 106(25): 3143-3421.

Barrett-Connor, E. (1991). "Nutrition epidemiology: how do we know what they ate?" The American journal of clinical nutrition 54(1 Suppl): 182S-187S.

Barrios, A., F. Blanco, et al. (2002). "Effect of dietary intervention on highly active antiretroviral therapy-related dyslipemia." AIDS 16(15): 2079-2081.

Booth, M. (2000). "Assessment of physical activity: an international perspective." Research quarterly for exercise and sport 71(2 Suppl): S114-120.

Brown, L., B. Rosner, et al. (1999). "Cholesterol-lowering effects of dietary fiber: a meta-analysis." The American journal of clinical nutrition 69(1): 30-42.

Bureau of Nutrition, D. o. H., Ministry of Public Health (2012). "Dietary Fiber Contents in Thai Foods."

Carr, A., K. Samaras, et al. (1999). "Diagnosis, prediction, and natural course of HIV-1 protease-inhibitor-associated lipodystrophy, hyperlipidaemia, and diabetes mellitus: a cohort study." Lancet 353(9170): 2093-2099.

Chuapai, Y., S. Kiertiburanakul, et al. (2007). "Lipodystrophy and dyslipidemia in human immunodeficiency virus-infected Thai patients receiving antiretroviral therapy." Journal of the Medical Association of Thailand = Chotmaihet thangphaet 90(3): 452-458.

Dube, M. P., D. Qian, et al. (2002). "Prospective, intensive study of metabolic changes associated with 48 weeks of amprenavir-based antiretroviral therapy." Clinical infectious diseases : an official publication of the Infectious Diseases Society of America 35(4): 475-481.

Feingold, K. R., R. M. Krauss, et al. (1993). "The hypertriglyceridemia of acquired immunodeficiency syndrome is associated with an increased prevalence of low density lipoprotein subclass pattern B." The Journal of clinical endocrinology and metabolism 76(6): 1423-1427.

Fellay, J., K. Boubaker, et al. (2001). "Prevalence of adverse events associated with potent antiretroviral treatment: Swiss HIV Cohort Study." Lancet 358(9290): 1322-1327.

Fitch, K. V., E. J. Anderson, et al. (2006). "Effects of a lifestyle modification program in HIV-infected patients with the metabolic syndrome." AIDS 20(14): 1843-1850.

Fitzmaurice Garrett M., L. N. M., Ware James H. (1962-). "Applied longitudinal analysis. 2nd ed. Hoboken, N.J. : Wiley, c2011."

Friis-Moller, N., C. A. Sabin, et al. (2003). "Combination antiretroviral therapy and the risk of myocardial infarction." The New England journal of medicine 349(21): 1993-2003.

Geil, P. B., J. W. Anderson, et al. (1995). "Women and men with hypercholesterolemia respond similarly to an American Heart Association step 1 diet." Journal of the American Dietetic Association 95(4): 436-441.

Greenberg, I., M. J. Stampfer, et al. (2009). "Adherence and success in long-term weight loss diets: the dietary intervention randomized controlled trial (DIRECT)." Journal of the American College of Nutrition 28(2): 159-168.

Grundy, S. M., J. I. Cleeman, et al. (2004). "Implications of recent clinical trials for the National Cholesterol Education Program Adult Treatment Panel III guidelines." Circulation 110(2): 227-239.

Grunfeld, C., M. Pang, et al. (1992). "Lipids, lipoproteins, triglyceride clearance, and cytokines in human immunodeficiency virus infection and the acquired immunodeficiency syndrome." The Journal of clinical endocrinology and metabolism 74(5): 1045-1052.

Henkin, Y., I. Shai, et al. (2000). "Dietary treatment of hypercholesterolemia: do dietitians do it better? A randomized, controlled trial." The American journal of medicine 109(7): 549-555.

Hiransuthikul, N., P. Hiransuthikul, et al. (2007). "Lipid profiles of Thai adult HIV-infected patients receiving protease inhibitors." The Southeast Asian journal of tropical medicine and public health 38(1): 69-77.

Hunninghake, D. B., E. A. Stein, et al. (1993). "The efficacy of intensive dietary therapy alone or combined with lovastatin in outpatients with hypercholesterolemia." The New England journal of medicine 328(17): 1213-1219.

Kathleen V. Fitch, E. J. A., Jane L. Hubbard, Sara J. Carpenter, William R. Waddell, Angela M. Caliendo, Steven K. Grinspoon (2006). "Effects of a lifestyle modification program in HIV-infected patients with the metabolic syndrome." AIDS 20: 8.

Kongkachuichai, R., R. Charoensiri, et al. (2010). "Carotenoid, flavonoid profiles and dietary fiber contents of fruits commonly consumed in Thailand." International journal of food sciences and nutrition 61(5): 536-548.

Kriska, A. M., W. C. Knowler, et al. (1990). "Development of questionnaire to examine relationship of physical activity and diabetes in Pima Indians." Diabetes care 13(4): 401-411.

Lazzaretti, R. K., R. Kuhmmer, et al. (2012). "Dietary intervention prevents dyslipidemia associated with highly active antiretroviral therapy in human immunodeficiency virus type 1-infected individuals: a randomized trial." Journal of the American College of Cardiology 59(11): 979-988.

Luara B. Almeida , A. C. S., Ana Clara F. Duran & Patricia C. Jaime (2011). "Impact of a nutritional counseling program on prevention of HAART-related metabolic and morphologic abnormalities." AIDS Care: Psychological and Socio-medical Aspects of AIDS/HIV 23(6): 9.

Moyle, G. J., M. Lloyd, et al. (2001). "Dietary advice with or without pravastatin for the management of hypercholesterolaemia associated with protease inhibitor therapy." AIDS 15(12): 1503-1508.

Ockene, I. S., J. R. Hebert, et al. (1999). "Effect of physician-delivered nutrition counseling training and an office-support program on saturated fat intake, weight, and serum lipid measurements in a hyperlipidemic population: Worcester Area Trial for Counseling in Hyperlipidemia (WATCH)." Archives of internal medicine 159(7): 725-731.

Palella, F. J., Jr., K. M. Delaney, et al. (1998). "Declining morbidity and mortality among patients with advanced human immunodeficiency virus infection. HIV Outpatient Study Investigators." The New England journal of medicine 338(13): 853-860.

Purnell, J. Q., A. Zambon, et al. (2000). "Effect of ritonavir on lipids and post-heparin lipase activities in normal subjects." AIDS 14(1): 51-57.

Rhodes, K. S., L. C. Bookstein, et al. (1996). "Intensive nutrition counseling enhances outcomes of National Cholesterol Education Program dietary therapy." Journal of the American Dietetic Association 96(10): 1003-1010; quiz 1011-1002.

Schambelan, M., C. A. Benson, et al. (2002). "Management of metabolic complications associated with antiretroviral therapy for HIV-1 infection: recommendations of an International AIDS Society-USA panel." Journal of acquired immune deficiency syndromes 31(3): 257-275.

Sever, P. S., B. Dahlof, et al. (2003). "Prevention of coronary and stroke events with atorvastatin in hypertensive patients who have average or lower-than-average cholesterol concentrations, in the Anglo-Scandinavian Cardiac Outcomes Trial--Lipid Lowering Arm (ASCOT-LLA): a multicentre randomised controlled trial." Lancet 361(9364): 1149-1158.

Shor-Posner, G., A. Basit, et al. (1993). "Hypocholesterolemia is associated with immune dysfunction in early human immunodeficiency virus-1 infection." The American journal of medicine 94(5): 515-519.

Sonia Maria de Figueiredo, M. G. M. G. P., Milena Maria Moreira Guimaraes, Lucas Jose de Campos Machado, Sidney Augusto Vieira Filho, Maria Arlene Fausto, et al. (2013). "Effects of dietary intervention on lipids profile of HIV infected patients on antiretroviral treatment (ART)." European Scientific Journal 9: 18.

Sprinz, E., R. K. Lazzaretti, et al. (2010). "Dyslipidemia in HIV-infected individuals." The Brazilian journal of infectious diseases : an official publication of the Brazilian Society of Infectious Diseases 14(6): 575-588.

Tsiodras, S., C. Mantzoros, et al. (2000). "Effects of protease inhibitors on hyperglycemia, hyperlipidemia, and lipodystrophy: a 5-year cohort study." Archives of internal medicine 160(13): 2050-2056.

Vergis, E. N., D. L. Paterson, et al. (2001). "Dyslipidaemia in HIV-infected patients: association with adherence to potent antiretroviral therapy." International journal of STD & AIDS 12(7): 463-468.

WHO (2004). "Nutrition Counselling Care and Support for HIV-infected Women: Guidelines on HIV-related care, treatment and support for HIV-infected women and their children in resource-contrained settings."

CHAPTER IV

Predictors of reducing low –density lipoprotein (LDL) cholesterol among Thai HIV-infected adults receiving a stable of antiretroviral regimen for at least 3 months

Abstract

Background: HIV-infected patients receiving antiretroviral therapy encounter the metabolic syndrome, including hyperlipidemia. LDL cholesterol is one of those significant predictors of cardiovascular disease (CVD) and it has been reported that reducing LDL-C is associated with an improvement of cardiovascular outcomes. The objective of this study was to identify predictors of reducing LDL-C in HIV-infected patients with abnormal LDL-C who are on stable antiretroviral therapy for at least 3 months.

Methods: We conducted a randomized, 24-week study in HIV-infected patients who were on antiretroviral therapy with dyslipidemia and were eligible to initiate therapeutic lifestyle changes (TLC) according to the National Cholesterol Education Program. Participants were randomly assigned into two groups. The intervention group received individual counseling with a nutritionist whereas the control group received standard care. All HIV positive patients with abnormal LDL-cholesterol were followed up for 24 weeks. Predictors (measured at baseline) associated with the reduction of LDL-C in HIV-infected patients were assessed for any reduction in LDL-C and for a reduction of at least 10 mg/dL.

Results: Seventy-two patients were randomly assigned and 64 (88.9%) participants completed the lipid profile tested at 24 weeks of the follow up. The odds of reducing LDL cholesterol level was

3.06 times (95%CI =1.06, 8.77) greater for those assigned to the group with individual nutritional counseling than those who were assigned to the standard care group. A reduction of LDL-C at any level and baseline LDL-C and the percentage of polyunsaturated fat (%PUFA) were significantly positively correlated. The percent PUFA was significant associated with LDL-C reduction at any level and of at least 10 mg/dL in the multivariate model. Similarly, a significant positive association was observed between good knowledge of dyslipidemia and LDL-C reduction of at least 10 mg/dL (adjusted OR =6.03, 95%CI = 1.40, 25.96).

Conclusions: The data demonstrated that knowledge of dyslipidemia is an important predictor for a reduction in LDL-C at least 10 mg/dL. This highlights the need for health professionals to provide adequate knowledge about dyslipidemia when treating HIV-infected patients with hyperlipidemia who are on antiretroviral drugs.

Keywords: LDL cholesterol reduction, antiretroviral therapy, HIV-infected adults, Thailand

Introduction

Dyslipidemia is one of the metabolic complications that have been increasingly reported in HIV-infected patients receiving antiretroviral therapy. The prevalence of dyslipidemia in HIV-infected individuals receiving antiretroviral therapy varies from 30% to 80% depending on drug combination and diagnostic definition criteria (Dube, Stein et al. 2003). Based on the Third Report of the National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (Adults Treatment Panel III), low-density lipoprotein (LDL) cholesterol has been identified as the primary target of cholesterol lowering therapy (2002; Grundy, Cleeman et al. 2004).

Low-density lipoprotein (LDL) cholesterol is a significant predictor of cardiovascular disease (CVD). A study (Howard, Robbins et al. 2000) showed that a 10 mg/dL increase in LDL-C was associated with a 12% increase in CVD risk. Several studies have shown that aggressively reducing LDL cholesterol lowers the incidence of CVD (Gordon, Kannel et al. 1981; Pyorala, Pedersen et al. 1997; Downs, Clearfield et al. 1998; Baigent, Keech et al. 2005). A recent study (Giraldez, Giugliano et al. 2008) found that baseline LDL cholesterol is an important predictor of the benefit of intensive lipid-lowering therapy compared with therapy of moderate intensity; a less intense effect of statin therapy was observed in patients with low baseline LDL-C. The study suggested that intensive lipid lowering therapy may not be necessary in patients whose baseline levels of LDL-C are already low.

Many previous studies have demonstrated reduction of cardiovascular events through intensive LDL-C reduction using lipid lowering medication. For every 1% reduction in LDL-C there is a corresponding 1-1.7% reduction in cardiovascular events. These studies indicated that the greater

LDL-C reduction, the larger decline in cardiovascular events risk (1995; Charland and Malone 2010; Li, Ambegaonkar et al. 2013). A reduction of LDL-C from baseline was associated with dosage of lipid lowering medication. A 6-week double blind clinical trial study showed a reduction of LDL-C levels by 25% - 61% with atorvastatin once a day with dosed of 2.5 mg to 80 mg (Nawrocki, Weiss et al. 1995). Other studies showed an association between LDL-C reduction and reduction in cardiovascular heart disease (CHD) events. Lowering LDL-C levels by 60 mg/dL reduced CHD event by 50% (Law, Wald et al. 2003). Similarly, a meta-analysis data of 90,056 HIV-negative persons in 14 randomized trials of statin therapy (Baigent, Keech et al. 2005) showed that a reduction of 1 mmol/L (about 39 mg/dL) in LDL-C levels lower rate of major coronary events by 23%.

Several factors have been shown to be associated with reducing LDL cholesterol. Dietary intervention has been suggested to be moderately effective for lowering lipid levels in HIV-infected patients with HAART-related dyslipidemia (Barrios, Blanco et al. 2002). Previous observational studies demonstrated that changes in weight were associated with changes in lipid levels and that total cholesterol and LDL-C levels declined with age in the elderly (Frishman, Ooi et al. 1992; Newschaffer, Bush et al. 1992; Wilson, Anderson et al. 1994). Male sex and older age were associated with LDL-C levels reduction in a cross-sectional study (Ferrara, Barrett-Connor et al. 1997). One study (Kumar P 2002) suggested women were more likely than men to develop an elevation of LDL-C levels when receiving some PI containing regimens. Pattibazel Geil's study (Geil, Anderson et al. 1995) showed that there was no difference between men and women in reduction of cholesterol levels after received an American Heart Association (AHA) step 1 diet (Energy from fat, carbohydrate and protein were 30%, 50% to 60% and 10% to 20%, respectively and cholesterol less than 300 mg per day) for 8 weeks.

To date, no study has been conducted to identify predictors of a reduction in LDL-C in Thai HIV-infected adults with dyslipidemia receiving antiretroviral therapy. It is well known that lowering LDL-C reduces coronary heart disease. Furthermore, there is evidence that HIV-infected patients receiving antiviral therapy are more likely to have dyslipidemia than those who are not on the antiretroviral therapy (Carr, Samaras et al. 1999; Purnell, Zambon et al. 2000; Tsiodras, Mantzoros et al. 2000; Fellay, Boubaker et al. 2001; Dube, Qian et al. 2002). Although lipid-lowering medications demonstrated effective reduction in LDL-C level, potential drug interaction between lipid-lowering medications and antiviral drugs in long-term is unknown. Lifestyle modification is recommended as the initial therapy for HIV-infected individuals with hyperlipidemia, similar to non HIV-infected persons. Thus, it is important to ascertain which factors can predict reduction in LDL-C levels among HIV-infected individuals receiving antiretroviral therapy in order to provide better care and management. The knowledge of these predictors will help clinicians to provide care and treatment for their HIV-infected patients. In addition, this will help policy makers to allocate a better plan for prevention and treatment of metabolic complications, particularly dyslipidemia. This study aims to identify predictors of reducing LDL-C in HIV-infected patients with abnormal LDL-C who have been on stable antiretroviral therapy for at least 3 months.

Methods

Study design and participants

We conducted a 24-week follow-up, randomized controlled study in HIV-infected patients who were on stable (at least 12 weeks) antiretroviral therapy with dyslipidemia and were eligible to initiate therapeutic lifestyle changes (TLC) according to the National Cholesterol Education Program (NCEP) (2002) (Schambelan, Benson et al. 2002). The aim of the study was to identify

predictors of reducing LDL cholesterol in HIV-infected patients who have started antiretroviral drug therapy and used the same regimen for at least three months. All HIV positive patients with abnormal LDL-cholesterol were followed up for 24 weeks. Predictors (measured at baseline) associated with the reduction of LDL-cholesterol in HIV-infected patients were assessed for any reduction in LDL-C and for a reduction of at least 10 mg/dL.

Data of this study was collected between March 2012 and March 2013. Participants were recruited from HIV-infected patients with abnormal LDL-cholesterol who were on antiretroviral therapy at least 3 months and visited the outpatient department (OPD) at Bamrasnaradura Hospital. Participants were screened for their lipid profile and risk of cardiovascular disease (CVD) according to NCEP. After the process of enrollment was completed, eighty eligible patients were recruited. Of these, eight refused to participate. The reasons for not participating were inconvenient to visit the hospital frequently or/and were unwilling to come if they did not have appointment with their own physicians. A total of 72 patients were randomized into two groups; 35 patients were in the individual counseling with a nutritionist group and 37 patients were in the standard care group. Thirteen withdrew and 59 completed the study at 24 weeks of follow up. Of these 29 (82.9%) and 30 (81.1%) were from intervention and control group, respectively (Fig.1).

Data collection

Prior to conduct interviews and measurements, informed consents were obtained from all eligible participants who were willing to participate in the study. At baseline, all participants had blood pressure, anthropometric data including weight, height, waist and hip circumference measured. Participants' dietary intake was assessed with 24-hr recall with trained nutritionists. After that,

participants were interviewed about their demographic characteristics, lifestyles behaviors; physical activity and alcohol consumption, and risk of cardiovascular diseases such as family history and smoking with self-administered questionnaires (see Appendix 2). Self–reported physical activity was assessed using the short interview version of the International Physical Activity Questionnaire (IPAQ) (Booth 2000). Physical activity level was calculated as the product of frequency and the duration of each activity in minutes per week, weighted by an estimate of the metabolic equivalent (MET) of the activity. The result was showed as the mean MET-min per week (Kriska, Knowler et al. 1990)

The participants in the control group received general verbal diet information at baseline and at subsequent annual visits as standard care, which is no individual counseling. Participants were assessed their dietary intake with a single 24-hr recall by a nutritionist at baseline and week 24 of the study.

The participants in the intervention group were given detailed advice on reducing saturated fat, trans fat, and cholesterol and increasing soluble fiber, including a Thai manual on nutritional promotion for HIV-infected patients with dyslipidemia (Chotivichien S 2012) and written information of Thai food based dietary guidelines (Bureau of Nutrition, MOPH). There was a total of 7 sessions with a professional nutritionist during the intervention: at baseline, 2 weeks, 4 weeks, 8 weeks, 12 weeks, 18 weeks and 24 weeks. The dietary intervention goals followed the Therapeutic Lifestyle Change (TLC) diet guidelines, which focus on LDL cholesterol changes according to the National Cholesterol Education Program (NCEP) Adult Treatment Panel III (ATP III). Food models were used to assess diet quantity. Participants were assessed for their dietary intake with 24-hr recall by a nutritionist at every visit. The dietary advice was be provided

based on participants' one week food records. Participants were asked to record food they consumed one week prior to the next visit (see Appendix 3). All participants received 200 Baht (~$7) for their participation at each visit. Lipid profile tests were collected and analyzed by technicians at the laboratory of Bamrasnaradura hospital. Cost for lipid profile tests at week 12 and 24 of all participants were paid by the study's fund. This study was reviewed and approved by Institutional Review Boards of the University of California at Los Angeles, the Thailand Ministry of Public Health Ethical Review Committee for Research in Human Subjects, and the Ethics Board of Bamrasnaradura hospital.

Measurements

Outcome variables

A reduction of LDL cholesterol (compared between baseline and 24 weeks of follow up) was divided into two "LDL reduction" categories by level of the reduction.

a. LDL cholesterol reduction **at any level** was classified as decrease at any level versus no change or increase.

b. LDL cholesterol reduction **at least 10 mg/dL** was classified as decrease at least 10 mg/dL versus no change or increase or decrease <10 mg/dL.

Participants were asked to fast 12 hours overnight before obtaining the venous blood on the next morning. Lipid profiles (total cholesterol, LDL cholesterol, HDL cholesterol and triglyceride) were measured using enzymatic colorimetric methods at the laboratory of Bamrasnaradura hospital.

Exposures and or covariates

Detailed information regarding these measurements is provided in chapter II (objectives and methods).

Data analyses

Descriptive analysis was used to describe qualitative variables. Each measurement and characteristic of interest at baseline was described as frequency distributions in terms of mean (and standard deviation) for continuous variables or number (percentages) for categorical variables. Differences between groups were compared using the two-sample t-test for continuous variables and Chi-square test or Fisher's exact tests for categorical variables. Nutrient intake, including energy, the amount of saturated fat (SFA), polyunsaturated fat (PUFA), monounsaturated fat (MUFA) was analyzed from dietary intake with the use of the Thai Fatty Acid program, using a Thai computerized dietary database. Pearson correlation were analyzed to verify bivariate interrelationships among LDL-C reduction, body mass index (BMI), waist, hip, percentage of saturated fatty (%SFA), monounsaturated fatty acid (%MUFA), polyunsaturated fatty acid (%PUFA), total fiber, soluble fiber and knowledge of dyslipidemia.

For simplification, we collapsed four responses of BMI into three categories: 0 = underweight, (<18.5 kg/m^2), 1= normal range (18·5–24·9 kg/ m^2) and 2=overweight or obese ($\geq$ 25 kg/m^2); six responses of education into two categories: 0 =less than University, 1= University or higher; five levels of income into 2 categories: 0 =15,000 Baht or below, 1= >15,000 Baht; four marital status into 2 categories: 0= single/never married, 1=married/ever married. For continuous variables: age was categorized into 3 age group: 18-34, 35-44 and 45 and above; duration of being known as HIV positive was categorized into two categories: 0=less than 10 years, 1= $\geq$10 years. In this study, we identified predictors of reducing LDL cholesterol in HIV-infected patients who received nutritional counseling or standard care, binary logistic regression was used. Best subset selection was used to obtain the best predictive model (Biostat406 class, A.Afifi, 2013).

$$\text{Logit}(R) = \alpha + \beta_1 X_1 + \beta_2 X_2 + \ldots + \beta_k X_k$$

R = Risk

α = Intercept

β = coefficient

X = exposures

All statistical analyses were conducted with SAS 9.1.3 statistical software package (SAS Institute Inc,. Cary, NC).

Results

Of these 72 participants, 64 (88.9%) completed lipid profile testing at baseline and 24 weeks. Baseline characteristics of those who completed lipid profile tests HIV-infected patients are illustrated in table 1. There was no significant difference in baseline characteristics between participants in the intervention group and the control group. Among 64 participants, 56.3% were female. The average age was 43.0 (standard deviation [s.d.] = 7.0). The average bodyweight was 60.2 kilograms (s.d.=10.9). The mean of body mass index (BMI) was 23.2 kg/m^2 (s.d.=3.5). The average waist circumference was 82.7 centimeters (s.d.=9.0). The average hip circumference was 93.9 centimeters (s.d.=6.7). In term of education, 18.8% had less than a high school education, 40.6% had graduated from high school or its equivalent and the remainder (40.6%) had graduated from University or higher. 12.5% had full time job either worked in private company or government. 23.4% worked part time, 46.9% were business owner and 18.2% were unemployed or a housewife. 21.9% reported a monthly income equal or less than 5,000 Baht. 57.7% reported a monthly income more than 10,000 Baht. Most of them (70.3%) were ever married. The majority of participants (80%) reported that they acquired HIV by engaging in heterosexual sex. One fourth of participants reported they had drunk alcohol in the past 30 days. 9 (14.1%) reported that they smoke cigarette. 17.2%, 96.9% and 85.9% of participants were receiving ARV with

protease, NRT and NNRT regimen, respectively. About one third of them had low level of knowledge about dyslipidemia.

There were no significant difference in baseline cardiovascular risk factors (including lipid profile, smoke cigarette, drink alcohol, hypertension and family history of metabolic diseases) and dietary parameters (including total energy, %Fat, %SFA, %PUFA %MUFA and cholesterol consumption) between participants in the intervention group and the control group. However, participants in the intervention group reported a slightly higher consumption of total fiber than participants in the control group (7.58 + 3.85 versus 5.83 + 3.00 g, P=0.047). Of 64 participants, approximately half of them (51.6%) had family history of at least one of metabolic disease or other such as diabetes (DM), hypertension (HT), cardiovascular disease and hyperlipidemia (Table 2).

Table 3 presents the proportion of LDL-C reduction in this study. The percentage of those in the invention group (individual counseling with a nutritionist) and the control group (without individual counseling) had a reduction in LDL cholesterol level at any level during the 24 weeks of follow up were 74% and 48%, respectively. For a reduction of LDL level of at least 10 mg/dL, the percentage of those in the invention group (individual counseling with a nutritionist) and the control group (without individual counseling) were 61% and 39%, respectively.

Table 4 illustrates the differences in knowledge of dyslipidemia between those who had decreased in LDL level and those who had not. Compared to those who did not succeed in reducing LDL level at least 10 mg/dL, better knowledge of dyslipidemia at baseline was observed in participants succeed in reducing LDL level at least 10 mg/dL (p=0.03). Bivariate correlations

among LDL reduction (at any level and at least 10 mg/dL), baseline LDL cholesterol, body mass index (BMI), waist, hip, saturated fatty acid (SFA), monounsaturated fatty acid (MUFA), polyunsaturated fatty acid (PUFA), total fiber, soluble fiber, and knowledge of dyslipidemia was showed in Table 5. LDL-C reduction at any level was positively correlated with baseline LDL cholesterol (r=0.26, p= 0.035) and %PUFA (r=0.31, p=0.012). A positive association between waist and hip circumference was observed (r=0.79, p=<0.01). BMI was positively correlated with both waist circumference (r=0.86, p=<0.001) and hip circumference (r=0.88, p=<0.001). There were positive association between %SFA and %PUFA (r=0.33, p=0.008) and between SFA and MUFA (r=0.88, p=<0.001). A negative association between SFA and total fiber was observed (r= -0.25, p=0.043). A reduction of LDL of at least 10 mg/dL was positively correlated with baseline LDL (r=0.80, p=<0.01), %PUFA (r=0.28, p=0.027) and knowledge of dyslipidemia (r=0.25, p=0.043).

The analyses to identify predictors for reducing LDL cholesterol (at any level) are presented in Table 6. The univariate analysis showed that the odds of reducing LDL cholesterol level was 3.06 times (95%CI =1.06, 8.77, p=0.04) greater for those assigned to the group with individual nutritional counseling than for those who was assigned to the standard care group. Baseline LDL cholesterol and percent PUFA were positively associated with LDL-C reduction (Crude OR =1.03, 95%CI = 1.001, 1.05 and Crude OR 1.61, 95%CI=1.07, 2.41). Table 7 presents univariate analysis examining the association between LDL-C reduction at least 10 mg/dL and potential predictors. In the univariate analysis, we found that participants with at least fair knowledge of dyslipidemia were more likely to achieve an LDL-C reduction of at least 10 mg/dL after 24 weeks of follow up (Crude OR=4.33, 95%CI=1.20, 15.69).

Variables obtained from best subset selection were included in the multivariate model. In the multivariate model, assignment to the intervention group (nutritional counseling) and a higher %PUFA consumption were positively associated with LDL-C reduction (adjusted OR =2.62, 95%CI = 0.87, 7.90) and (adjusted OR=1.55, 95%CI= 1.03, 2.33), respectively (table 8). Table 9 presents multivariate analysis examining the association between LDL-C reduction at least 10 mg/dL and potential predictors. We found that participants who answered 6-7 questions (out of 10) about dyslipidemia in the questionnaire correctly were more likely to experience an LDL-C reduction at least 10 mg/dL at 24 weeks of follow up (adjusted OR =5.14, 95%CI = 1.25, 21.21). Similarly, participants who answered 8-10 questions correctly (good knowledge about dyslipidemia) reduced their LDL level at least 10 mg/dL (adjusted OR =6.03, 95%CI = 1.40, 25.96). Lastly, a significant positive association between %PUFA and reduction of LDL-C at least 10 mg/dL was observed (adjusted OR =1.50, 95%CI= 1.05, 2.14).

Discussion

It is well known that lowering LDL-C reduces the risk of coronary heart disease. Furthermore, there is evidence that HIV-infected patients receiving antiviral therapy are more likely to have dyslipidemia, including elevated LDL-C than those who are not on the antiviral therapy. To our knowledge, no study to identify the predictors for a reduction of low-density lipoprotein (LDL) cholesterol in Thai HIV-infected persons has been reported. Previous studies in western countries have demonstrated a reduction of LDL-C through the use of lipid-lowering medication. The present study is one of the first to identify those predictors and their associations with a reduction of LDL-C in HIV-infected patients receiving antiretroviral therapy but not currently taking lipid-lowering medication.

Our study is consistent with other studies that counseling effectively improved lipid and other risk cardiovascular factors in patients not receiving lipid-lowering drugs (Ockene, Hebert et al. 1999; Henkin, Shai et al. 2000). The present study showed higher proportions of patients reduced their LDL-C in the intervention and control groups (74% vs. 48%) than in Henkin's study (Henkin, Shai et al. 2000), which found a decrease of LDL-C level of 65% and 35% in the dietitian group and the physician group, respectively. Based on our study, none of the participants' demographic characteristics (age, gender, body mass index, education, occupation, income and marital status) were associated with lowering LDL-C levels. This finding is inconsistent with some studies that showed higher LDL-C reduction with older age (Frishman, Ooi et al. 1992; Newschaffer, Bush et al. 1992), male sex (Ferrara, Barrett-Connor et al. 1997) and obesity (Wilson, Anderson et al. 1994). However, our study was conducted in a younger population and less obese population (most prior studies were conducted in an elderly population).

Our study demonstrated that baseline LDL-C was positively associated with LDL-C reduction. This finding was in agreement with previous studies of LDL-C reduction using lipid-lowering medications (Holme 1995; Giraldez, Giugliano et al. 2008). The predictors associated with a reduction of LDL-C at any level were assignment to the intervention group (adjusted OR= 2.62, 95%CI= 0.87, 7.90) and higher percentages of polyunsaturated fatty acid (% PUFA) consumption (adjusted OR= 1.55, 95%CI = 1.03, 2.33).

Our finding demonstrated that percentage of PUFA is a predictor of reducing LDL-C. Participants who consumed greater %PUFA from baseline tended to lower LDL at 24 weeks of follow up more than those who consumed lesser. This finding is consistent with Mattson and

Grundy's study (Mattson and Grundy 1985) that showed a significant reduction of LDL-C in participants who consumed unsaturated fatty acid diets, both monounsaturated and polyunsaturated fatty acids, compared to those who consumed saturated fatty acid diet.

While one prospective study (Barrios, Blanco et al. 2002) showed a higher reduction in cholesterol levels in participants treated with PI-based combinations of antiretroviral therapy, our study did not observe an association between treatment with PI-based and reduction of LDL-C. Our study demonstrated that assignment into the intervention group (nutritional counseling) was associated with LDL reduction. This finding is consistent with several previous studies that used dietary modification i.e. an increase in the polyunsaturated to saturated fat ratio in the diet and an increase in soluble fiber intake showed modest effect in reducing LDL-C (Tang, Armitage et al. 1998; Brown, Rosner et al. 1999). Patients with higher %PUFA and who had a good level of knowledge about dyslipidemia were more likely to attain reduction in LDL-C at 24 weeks of follow up. The multivariate analysis showed that at least fair knowledge of dyslipidemia was a predictor for achieving LDL-C reduction of at least 10 mg/dL. A previous randomized study (Shannon, Tershakovec et al. 1994) had also demonstrated that high dietary knowledge in parents was associated with a significantly decline in mean LDL-C levels of their children. Our study contrasts with some study (Hadigan, Jeste et al. 2001) in that we did not observe an association between alcohol consumption and LDL-C levels. Our finding demonstrated an effect of polyunsaturated fatty acids (PUFAs) in reducing LDL-C. This finding is consistent with results of a meta-analysis from 10 randomized controlled trials with 557 end-stage renal diseases patients (Pei, Zhao et al. 2012) that showed a negative association between n-3 PUFA consumption and LDL-C level. Similarly, a recent prospective study in 2,692 adults in 4 U.S. communities showed that high blood levels of long chain ω3-PUFA were associated with lower coronary heart disease

(CHD) death in the elderly. This was similar to a negative association between LDL-C levels and CHD (Mozaffarian D 2013).

A prospective study (Barrios, Blanco et al. 2002) showed that reductions in cholesterol levels were more substantial in participants treated with PI-based combinations of antiretroviral drugs. In contrast, our study did not observe an association between treatment with PI-based and reduction of LDL-C. Although many studies (Henry, Melroe et al. 1998; Moyle, Lloyd et al. 2001; Barrios, Blanco et al. 2002; Batterham, Brown et al. 2003) had demonstrated that diet changes, particularly fat intake ameliorated dyslipidemia among HIV-infected patients receiving antiretroviral drugs, our study did not find an association between fat intake and LDL-C reduction.

Some limitations in our study should be noted. First, a small sample size may lead to low power to detect an association in some parameters such as an association of using PIs and lowering LDL-C and we did not have proper sample size to analyze data separately in the assignment groups (nutritional counseling vs control) as we proposed. Secondly, due to the lack of a database of soluble fiber content in Thai food, the present study could not demonstrate the benefit of soluble fiber on LDL-C reduction. Lastly, we used only baseline data to analyze the results, thus we did not demonstrate an association of weight loss and a reduction of LDL-C in this study.

Based on our study, a predictor for achieving a reduction of LDL-C at any level at 24 weeks of follow up was a high percentage of PUFA consumption. Higher baseline LDL-C and assigned to the nutritional counseling group were also led to a reduction of LDL-C levels. We found that higher baseline LDL-C was positively associated with greater reductions in LDL-C although it

was not significant in the multivariate analysis. Knowledge of dyslipidemia was demonstrated to be a strong predictor for a reduction of LDL-C at least 10 mg/dL. Participants with high baseline LDL-C levels that were still in the range of therapeutic lifestyle changes (TLC) tended to show decreased LDL-C after 24 weeks of follow up. Thus, physicians who care for HIV-infected patients with high baseline LDL-C that remain in the range of TLC should not immediately start lipid-lowering drugs before allowing time (at least 24 weeks) for patients to adjust their lifestyle and knowledge of dyslipidemia should be provided to every dyslipidemia patient.

In conclusion, our study underscored the importance of lowering LDL-C levels through lifestyle modification without using lipid-lowering medication. Individualized nutrition counseling, diet modification, substitution of SFA with PUFA, and knowledge of dyslipidemia should be emphasized in lifestyle changes of HIV-infected adults receiving antiretroviral therapy with hyperlipidemia.

Table 1. Baseline characteristics of participants who completed lipid profile tests at 24 weeks of follow up

Characteristics	Total (N=64) Mean (s.d.*); N (%)	Intervention group (N=31) Mean (s.d.*); N (%)	Control group (N=33) Mean (s.d.*); N (%)	p-value
Age (years)	43.0 (7.0)	42.1 (6.4)	43.9 (7.6)	0.316
Female gender – N (%)	36 (56.3%)	20 (64.5%)	16 (48.5%)	0.196
Anthropometric measurement				
Bodyweight (kgs)	60.2 (10.9)	58.4 (9.0)	61.9 (12.3)	0.211
Body Mass Index (kg/m^2)	23.2 (3.5)	23.1 (2.9)	23.3 (4.1)	0.833
Waist circumference (cm)	82.7 (9.0)	81.7 (7.6)	83.7 (10.1)	0.369
Hip circumference (cm)	93.9 (6.7)	93.2 (4.9)	94.5 (8.0)	0.422
Waist hip ratio	0.88 (0.06)	0.88 (0.06)	0.88 (0.06)	0.565
Education – N (%)				0.484
Less than a high school	12 (18.8%)	6 (19.4%)	6 (18.2%)	
High school or its equivalent	26 (40.6%)	15 (48.4%)	11 (33.3%)	
University or higher	26 (40.6%)	10 (32.3%)	16 (48.5%)	
Occupation – N (%)				0.365
Full time: Government or Private	8 (12.5%)	3 (9.7%)	5 (15.2%)	
Part time: Regular or irregular	15 (23.4%)	8 (25.8%)	7 (21.2%)	
Owner of business	26 (40.6%)	12 (38.7%)	18 (54.6%)	
Unemployed	11 (17.2%)	8 (25.8%)	3 (9.1%)	
Married/ever married – N (%)	45 (70.3%)	24 (77.4%)	21 (63.6%)	0.602

*s.d. = Standard Deviation

Table 1. (cont.) Baseline characteristics of participants who completed lipid profile tests at 24 weeks of follow up

Characteristics	Total (N=64) Mean (s.d.*); N (%)	Intervention group (N=31) Mean (s.d.*); N (%)	Control group (N=33) Mean (s.d.*); N (%)	p-value
Income (Baht) – N (%)				0.361
5,000 or less	14 (21.9%)	10 (32.3%)	4 (12.1%)	
>5,000 – 10,000	13 (20.3%)	6 (19.4%)	7 (21.2%)	
>10,000	37 (57.8%)	15 (48.4%)	22 (66.7%)	
Duration of being known as HIV positive (years)	9.75 (4.6)	10.16 (5.2)	9.36 (4.0)	0.495
Route of HIV acquisition – N (%)				0.554
Heterosexual sex	51 (79.7%)	24 (77.4%)	27 (81.8%)	
Homosexual sex	8 (12.5%)	5 (16.1%)	3 (9.1%)	
Intravenous drug	1 (1.6%)	1 (3.2%)	0 (0.0)	
Bisexual sex	2 (3.1%)	0 (0.0)	2 (6.1%)	
Tattoo	2 (3.1%)	1 (3.2%)	1 (3.0%)	
Current ARV treatment – N (%)				
Treatment with protease	11 (17.2%)	4 (12.9%)	7 (21.2%)	0.512
Treatment with NRT	62 (96.9%)	30 (96.8%)	32 (97.0%)	1.000
Treatment with NNRT	55 (85.9%)	27 (87.1%)	28 (84.9%)	1.000
Knowledge of dyslipidemia – N (%)				0.565
Poor	22 (34.4)	9 (29.0%)	13 (39.4%)	
Fair	21 (32.8)	10 (32.3%)	11 (33.3%)	
Good	21 (32.8)	12 (38.7%)	9 (27.3%)	

*s.d. = Standard Deviation

Table 2. Baseline cardiovascular risk factors and diet of participants who completed lipid profile tests at 24 weeks of follow up

Characteristics	Total (N=64) Mean (s.d.*); N (%)	Intervention group (N=31) Mean (s.d.*); N (%)	Control group (N=33) Mean (s.d.*); N (%)	p-value
Cardiovascular risk factors – N (%)				
Total Cholesterol (mg/dL)	229.5 (26.5)	232.4 (30.5)	226.8 (22.3)	0.401
LDL-C (mg/dL)	159.9 (23.6)	163.2 (24.5)	156.7 (22.6)	0.271
HDL-C (mg/dL)	56.6 (17.4)	56.6 (18.3)	56.6 (16.7)	0.995
Triglyceride (mg/dL)	160.5 (95.3)	159.2 (92.3)	161.8 (99.5)	0.914
Smoke cigarette	9 (14.1%)	4 (12.9%)	5 (15.2%)	1.000
Drink alcohol	16 (25.0%)	9 (29.0%)	7 (21.2%)	0.568
Treatment with hypertension	5 (7.8%)	2 (6.5%)	3 (9.1%)	1.000
Family history of metabolic diseases	33 (51.6%)	19 (61.3%)	14 (42.4%)	0.131
Dietary parameters				
Energy (kcal)	1365.6 (459.8)	1402.8 (535.0)	1330.7 (381.3)	0.535
Fat (%)	27.3 (8.4)	26.9 (9.5)	27.7 (7.4)	0.701
Saturated fat (%)	5.0 (4.6)	5.0 (4.0)	5.0 (5.2)	0.958
Polyunsaturated fat (%)	2.0 (1.8)	2.3 (2.0)	1.7 (1.7)	0.164
Monounsaturated fat (%)	4.5 (3.7)	4.7 (4.0)	4.3 (3.5)	0.661
Cholesterol (mg)	187.0 (144.5)	163.2 (131.0)	209.3 (154.7)	0.205
Total fiber (g)	6.7 (3.5)	7.6 (3.9)	5.8 (3.0)	0.047

*s.d. = Standard Deviation

Table 3. Number of HIV-infected adults who had LDL-C reduction at any level and at least 10 mg/dL at 24 weeks of follow-up, by group (n=64)

Group	LDL-C reduction at any level*		LDL-C reduction at least 10 mg/dL**	
	Yes (%)	No (%)	Yes (%)	No (%)
Intervention	23 (74.2)	8 (25.8)	19 (61.3)	12 (38.7)
Control	16 (48.5)	17 (51.5)	13 (39.4)	20 (60.6)
Total	39 (60.9)	25 (39.1)	32 (50.00)	32 (50.00)

*p=0.04
**p=0.08

Table 4. Knowledge of dyslipidemia among HIV-infected adults according to LDL-C reduction status

Knowledge of dyslipidemia	LDL-C reduction at any level*		LDL-C reduction at least 10 mg/dL**	
	Yes (%)	No (%)	Yes (%)	No (%)
Good	14 (21.9)	7 (10.9)	13 (20.3)	8 (12.5)
Fair	14 (21.9)	7 (10.9)	13 (20.3)	8 (12.5)
Poor	11 (17.2)	11 (17.2)	6 (9.4)	16 (25.0)
Total	39 (60.9)	25 (39.1)	32 (50.0)	32 (50.0)

*p = 0.43
**p =0.03

Table 5. Bivariate correlation among variables of interest

Variables	1	2	3	4	5	6	7	8	9	10	11	12
1. LDL reduction (At any level)	1											
2. Baseline LDL-C	0.26*	1										
3. BMI	0.14	0.21	1									
4. Waist	0.01	0.18	0.86**	1								
5. Hip	0.08	0.12	0.88**	0.79**	1							
6. SFA (%)	-0.06	0.09	-0.04	-0.05	-0.07	1						
7. PUFA (%)	0.31*	0.30*	0.10	0.002	0.17	0.33*	1					
8. MUFA (%)	0.05	0.15	-0.09	-0.13	-0.09	0.88**	0.53**	1				
9. Total fiber	0.04	-0.20	0.03	-0.02	-0.03	-0.25*	-0.07	-0.20	1			
10. Soluble fiber	0.12	-0.007	0.17	0.10	0.23	-0.11	0.02	-0.14	0.20	1		
11. Knowledge of dyslipidemia	0.14	0.11	-0.04	0.01	-0.04	-0.20	-0.11	-0.22	-0.06	0.11	1	
12. LDL reduction (At least 10 mg/dL)	0.80**	0.24*	0.19	0.05	0.11	-0.05	0.28*	0.09	-0.02	0.17	0.25*	1

*p<0.05
**p<0.001

Table 6. Univariate analysis of related factors and reducing LDL cholesterol at any level among HIV-infected patients

Variables	Crude OR	95%CI	*p*
Age group (year)			
18-34	1.00		
35-44	0.59	0.06, 6.27	0.66
45 or above	0.39	0.04, 4.35	0.45
Group (intervention)	3.06	1.06, 8.77	**0.04**
Gender (male)	0.44	0.16,1.23	0.12
BMI: Underweight	1.00		
Normal	0.42	0.04, 4.26	0.46
Overweight or obese	1.11	0.08, 15.04	0.94
Education (≥ University)	0.80	0.29, 2.21	0.66
Income (> 15,000 Baht)	0.52	0.18, 1.52	0.23
Marital status (ever married)	1.20	0.40, 3.57	0.75
Occupation: Unemployed	1.00		
Full time job	0.95	0.14, 6.28	0.96
Part time job	1.57	0.29, 8.42	0.60
Owner of business	0.65	0.16, 2.71	0.56
Smoke cigarette	0.77	0.19, 3.20	0.72
Drink alcohol	1.09	0.34, 3.50	0.88
Treatment with Protease	0.73	0.20, 2.70	0.63
Knowledge of dyslipidemia			
Poor	1.00		
Fair	2.00	0.58, 6.87	0.27
Good	2.00	0.58, 6.87	0.27
Duration of being known as HIV positive (≥ 10 years)	0.98	0.36, 2.71	0.97
Baseline LDL-C (mg/dL)	1.03	1.001,1.05	**0.04**
PUFA (% of total calories)	1.61	1.07, 2.41	**0.02**

Table 7. Univariate analysis of related factors and reducing LDL cholesterol at least 10 mg/dL among HIV-infected patients

Variables	Crude OR	95%CI	*p*
Age group (year)			
18-34	1.00		
35-44	0.42	0.04, 4.40	0.47
45 or above	0.20	0.02, 2.23	0.19
Group (intervention)	2.44	0.89, 6.65	0.08
Gender (male)	0.60	0.22, 1.63	0.31
BMI: Underweight	1.00		
Normal	0.27	0.03, 2.78	0.27
Overweight or obese	0.53	0.04, 6.66	0.63
Education (≥ University)	1.00	0.37, 2.71	1.00
Income (> 15,000 Baht)	0.75	0.26, 2.16	0.59
Marital status (ever married)	0.64	0.22, 1.88	0.41
Occupation: Unemployed	1.00		
Full time job	0.83	0.13, 5.17	0.84
Part time job	1.67	0.34, 8.26	0.53
Owner of business	0.56	0.14, 2.24	0.41
Smoke cigarette	0.77	0.19, 3.18	0.72
Drink alcohol	1.00	0.32, 3.10	1.00
Treatment with Protease	0.31	0.07, 1.31	0.09
Knowledge of dyslipidemia			
Poor	1.00		
Fair	4.33	1.20, 15.69	**0.03**
Good	4.33	1.20, 15.69	**0.03**
Duration of being known as HIV positive (≥ 10 years)	1.00	0.37, 2.69	1.00
Baseline LDL-C (mg/dL)	1.02	1.00, 1.05	0.06
PUFA (% of total calories)	1.41	1.02, 1.94	**0.04**

Table 8. Multivariate analysis of related factors and reducing LDL cholesterol at any level among HIV-infected patients

Variables	Adjusted OR	95%CI	*p*
Group (intervention)	2.62	0.87, 7.90	0.09
PUFA (% of total calories)	1.55	1.03, 2.33	**0.04**

Table 9. Multivariate analysis of related factors and reducing LDL cholesterol at least 10 mg/dL among HIV-infected patients

Variables	Adjusted OR	95%CI	*p*
Group (intervention)	1.97	0.65, 6.01	0.23
Knowledge of dyslipidemia			
Poor	1.00		
Fair	5.14	1.25, 21.21	**0.02**
Good	6.03	1.40, 25.96	**0.01**
PUFA (% of total calories)	1.50	1.05, 2.14	**0.03**

References

(1995). "Baseline serum cholesterol and treatment effect in the Scandinavian Simvastatin Survival Study (4S)." Lancet 345(8960): 1274-1275.

(2002). "Third Report of the National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel III) final report." Circulation 106(25): 3143-3421.

Afifi, A. A. (2013) Biostat 406 class, retrieved on <http://www.biostat.ucla.edu/course/406/Biostat406-LogisticRegression2.pdf>.

Baigent, C., A. Keech, et al. (2005). "Efficacy and safety of cholesterol-lowering treatment: prospective meta-analysis of data from 90,056 participants in 14 randomised trials of statins." Lancet 366(9493): 1267-1278.

Barrios, A., F. Blanco, et al. (2002). "Effect of dietary intervention on highly active antiretroviral therapy-related dyslipemia." AIDS 16(15): 2079-2081.

Batterham, M. J., D. Brown, et al. (2003). "Modifying dietary fat intake can reduce serum cholesterol in HIV-associated hypercholesterolemia." AIDS 17(9): 1414-1416.

Brown, L., B. Rosner, et al. (1999). "Cholesterol-lowering effects of dietary fiber: a meta-analysis." The American journal of clinical nutrition 69(1): 30-42.

Bursac, Z., C. H. Gauss, et al. (2008). "Purposeful selection of variables in logistic regression." Source code for biology and medicine 3: 17.

Carr, A., K. Samaras, et al. (1999). "Diagnosis, prediction, and natural course of HIV-1 protease-inhibitor-associated lipodystrophy, hyperlipidaemia, and diabetes mellitus: a cohort study." Lancet 353(9170): 2093-2099.

Charland, S. L. and D. C. Malone (2010). "Prediction of cardiovascular event risk reduction from lipid changes associated with high potency dyslipidemia therapy." Current medical research and opinion 26(2): 365-375.

Downs, J. R., M. Clearfield, et al. (1998). "Primary prevention of acute coronary events with lovastatin in men and women with average cholesterol levels: results of AFCAPS/TexCAPS. Air Force/Texas Coronary Atherosclerosis Prevention Study." JAMA : the journal of the American Medical Association 279(20): 1615-1622.

Dube, M. P., D. Qian, et al. (2002). "Prospective, intensive study of metabolic changes associated with 48 weeks of amprenavir-based antiretroviral therapy." Clinical infectious diseases : an official publication of the Infectious Diseases Society of America 35(4): 475-481.

Dube, M. P., J. H. Stein, et al. (2003). "Guidelines for the evaluation and management of dyslipidemia in human immunodeficiency virus (HIV)-infected adults receiving antiretroviral therapy: recommendations of the HIV Medical Association of the Infectious Disease Society of America and the Adult AIDS Clinical Trials Group." Clinical infectious diseases : an official publication of the Infectious Diseases Society of America 37(5): 613-627.

Fellay, J., K. Boubaker, et al. (2001). "Prevalence of adverse events associated with potent antiretroviral treatment: Swiss HIV Cohort Study." Lancet 358(9290): 1322-1327.

Ferrara, A., E. Barrett-Connor, et al. (1997). "Total, LDL, and HDL cholesterol decrease with age in older men and women. The Rancho Bernardo Study 1984-1994." Circulation 96(1): 37-43.

Frishman, W. H., W. L. Ooi, et al. (1992). "Serum lipids and lipoproteins in advanced age. Intraindividual changes." Annals of epidemiology 2(1-2): 43-50.

Geil, P. B., J. W. Anderson, et al. (1995). "Women and men with hypercholesterolemia respond similarly to an American Heart Association step 1 diet." Journal of the American Dietetic Association 95(4): 436-441.

Giraldez, R. R., R. P. Giugliano, et al. (2008). "Baseline low-density lipoprotein cholesterol is an important predictor of the benefit of intensive lipid-lowering therapy: a PROVE IT-TIMI 22 (Pravastatin or Atorvastatin Evaluation and Infection Therapy-Thrombolysis In Myocardial Infarction 22) analysis." Journal of the American College of Cardiology 52(11): 914-920.

Gordon, T., W. B. Kannel, et al. (1981). "Lipoproteins, cardiovascular disease, and death. The Framingham study." Archives of internal medicine 141(9): 1128-1131.

Grundy, S. M., J. I. Cleeman, et al. (2004). "Implications of recent clinical trials for the National Cholesterol Education Program Adult Treatment Panel III guidelines." Circulation 110(2): 227-239.

Hadigan, C., S. Jeste, et al. (2001). "Modifiable dietary habits and their relation to metabolic abnormalities in men and women with human immunodeficiency virus infection and fat redistribution." Clinical infectious diseases : an official publication of the Infectious Diseases Society of America 33(5): 710-717.

Henkin, Y., I. Shai, et al. (2000). "Dietary treatment of hypercholesterolemia: do dietitians do it better? A randomized, controlled trial." The American journal of medicine 109(7): 549-555.

Henry, K., H. Melroe, et al. (1998). "Atorvastatin and gemfibrozil for protease-inhibitor-related lipid abnormalities." Lancet 352(9133): 1031-1032.

Holme, I. (1995). "Cholesterol reduction and its impact on coronary artery disease and total mortality." The American journal of cardiology 76(9): 10C-17C.

Howard, B. V., D. C. Robbins, et al. (2000). "LDL cholesterol as a strong predictor of coronary heart disease in diabetic individuals with insulin resistance and low LDL: The Strong Heart Study." Arteriosclerosis, thrombosis, and vascular biology 20(3): 830-835.

Kumar P, R.-F. A., Thompson M, et al. (2002). Prospective study of hyperlipidemia in ART-naïve subjects taking combivir/abacavir (COM/ABC), COM/nelfinavir (NFV), or stavudine (d4T)/lamivudine (3TC)/NFV (ESS40002) [abstract 33]. the 9th Conference on Retroviruses and Opportunistic Infections Seattle.

Law, M. R., N. J. Wald, et al. (2003). "Quantifying effect of statins on low density lipoprotein cholesterol, ischaemic heart disease, and stroke: systematic review and meta-analysis." BMJ 326(7404): 1423.

Li, L., B. M. Ambegaonkar, et al. (2013). "Association of a reduction in low-density lipoprotein cholesterol with incident cardiovascular and cerebrovascular events among people with type 2 diabetes mellitus." European journal of preventive cardiology.

Mattson, F. H. and S. M. Grundy (1985). "Comparison of effects of dietary saturated, monounsaturated, and polyunsaturated fatty acids on plasma lipids and lipoproteins in man." Journal of lipid research **26**(2): 194-202.

Moyle, G. J., M. Lloyd, et al. (2001). "Dietary advice with or without pravastatin for the management of hypercholesterolaemia associated with protease inhibitor therapy." AIDS 15(12): 1503-1508.

Mozaffarian D, L. R., King IB, Song X., Huang H., Sacks FM, Rimm EB., Wang M., Siscovick DS. (2013). "Low blood level of W3 PUFA as strong predictor of CHD as high LDLcholesterol." Ann Intern Med 158: epub.

Nawrocki, J. W., S. R. Weiss, et al. (1995). "Reduction of LDL cholesterol by 25% to 60% in patients with primary hypercholesterolemia by atorvastatin, a new HMG-CoA reductase inhibitor." Arteriosclerosis, thrombosis, and vascular biology 15(5): 678-682.

Newschaffer, C. J., T. L. Bush, et al. (1992). "Aging and total cholesterol levels: cohort, period, and survivorship effects." American journal of epidemiology 136(1): 23-34.

Ockene, I. S., J. R. Hebert, et al. (1999). "Effect of physician-delivered nutrition counseling training and an office-support program on saturated fat intake, weight, and serum lipid measurements in a hyperlipidemic population: Worcester Area Trial for Counseling in Hyperlipidemia (WATCH)." Archives of internal medicine 159(7): 725-731.

Pei, J., Y. Zhao, et al. (2012). "The effect of n-3 polyunsaturated fatty acids on plasma lipids and lipoproteins in patients with chronic renal failure--a meta-analysis of randomized controlled trials." Journal of renal nutrition : the official journal of the Council on Renal Nutrition of the National Kidney Foundation 22(6): 525-532.

Purnell, J. Q., A. Zambon, et al. (2000). "Effect of ritonavir on lipids and post-heparin lipase activities in normal subjects." AIDS 14(1): 51-57.

Pyorala, K., T. R. Pedersen, et al. (1997). "Cholesterol lowering with simvastatin improves prognosis of diabetic patients with coronary heart disease. A subgroup analysis of the Scandinavian Simvastatin Survival Study (4S)." Diabetes care 20(4): 614-620.

Shannon, B. M., A. M. Tershakovec, et al. (1994). "Reduction of elevated LDL-cholesterol levels of 4- to 10-year-old children through home-based dietary education." Pediatrics 94(6 Pt 1): 923-927.

Tang, J. L., J. M. Armitage, et al. (1998). "Systematic review of dietary intervention trials to lower blood total cholesterol in free-living subjects." BMJ 316(7139): 1213-1220.

Tsiodras, S., C. Mantzoros, et al. (2000). "Effects of protease inhibitors on hyperglycemia, hyperlipidemia, and lipodystrophy: a 5-year cohort study." Archives of internal medicine 160(13): 2050-2056.

Wilson, P. W., K. M. Anderson, et al. (1994). "Determinants of change in total cholesterol and HDL-C with age: the Framingham Study." Journal of gerontology 49(6): M252-257.

CHAPTER V

SUMMARY

HIV/AIDS remains one of the most important public health problems in Thailand. In 2010, Thailand was one of the countries in the Southeast Asian region that reported adult HIV prevalence over 1% (WHO-SEAR 2012). Nearly 1 in 75 people are infected with HIV and over 200,000 people are under treatment for HIV infection with antiretroviral therapy (Bureau of Epidemiology). The current accessibility and availability of antiretroviral therapy and earlier initiation of therapy make the metabolic complications from antiretroviral therapy inevitable. Proper nutrition is important not only to improve the quality of life of HIV-infected persons by selecting proper foods (both quantity and quality) early in infection, but also to help reduce drug and food interactions. While nutritional interventions to prevent metabolic complications of HIV and antiretroviral therapy have been studied elsewhere, none have been reported from Thailand, where dietary patterns and population genetics differ from those in other regions.

Our study was the first to describe the effect of individual nutritional counseling on low density lipoprotein (LDL) cholesterol reduction in HIV-infected persons receiving antiretroviral therapy in Thailand. In addition, it explored the possible ways to provide nutritional sessions for HIV-infected patients treated in Thai settings. Our findings emphasize the importance of nutrition counseling in controlling dyslipidemia and reducing metabolic complications in HIV-infected patients receiving antiretroviral therapy. Our findings support that individual nutritional counseling helps patients to reduce their total cholesterol, particularly LDL cholesterol. Based on the study, an intensive nutritional counseling intervention was feasible for patients and resulted in favorable outcomes. We speculate that patients might be more adherent with an aggressive

nutritional counseling schedule if appointments were coordinated with appointments with their own physicians and if their own physician prescribed nutritional counseling.

This present study demonstrated a desirable outcome of LDL-C reduction in the group receiving individual nutritional counseling but not in the standard care group. However, this finding is not solely explained by adherence to the dietary intervention recommendations for specific macronutrients. A reduction of fat and cholesterol intake was reported in both groups. Surprisingly, a greater percentage of individuals in the control group than the intervention group reported adherence to the dietary recommendation at 24 weeks regarding percent fat, SFA, PUFA and MUFA. The intervention group did however reduce their total caloric intakes and their weight. Although our findings did not demonstrated that participants in the intervention group met specific dietary recommendations, we did observe desirable changes in percent calories from protein and in total carbohydrate intake. The weight loss and total energy reduction in the intervention group in this study might partial explain their LDL-C reduction.

Despite the small sample size and apparent non-adherence to macronutrient recommendations, our study demonstrated improvement with individual nutritional counseling in lipid profiles in HIV-positive persons antiretroviral therapy. These participants who received nutritional counseling showed an improvement in lipid profile that was not observed in the control group. We suspect the major contributions to the effect derived from a reduction in weight in most preobese and obese participants. While the intervention group demonstrated a significant improvement in lipid profiles, particularly LDL-C and total cholesterol, our findings failed to demonstrate increased adherence to the dietary recommendations among participants who received individual nutritional counseling. Our failure to observe the desirable dietary adherences may partially be explained by

the limitation of using a single 24 hour recall to obtain the percentage of participants who adhered to the dietary recommendations. This method causes biased estimates of the percentage of adherence because of a high variability of people's day-to-day dietary intake (CDC 2013). We used a single 24 hour recall because it is easy and practical to conduct in Thai settings and it gives unbiased estimate of the mean dietary intake for the group. It does not provide a stable individual mean intake and this variance leads to lower detection of significant differences in intake. In the present study, nutritional counseling demonstrated an effect on LDL-C and total cholesterol reduction but we could not show that participants in the intervention group adhered to the dietary recommendations better than those in the control group. The limitation of the inflated variance from a single 24 hour recall should be kept in mind when interpreting the adherence data.

Our findings emphasized the importance of integrating individual nutritional counseling in the HIV clinic. If patients' dyslipidemia can be treated with lifestyle modification, this will lead to lower costs of treatment and prevent drug interactions that might occur between lipid lowering medication and antiretroviral therapy in long-term treatment. Moreover, our study demonstrated an important implication for HIV/AIDS treatment and care, and also for future research that addresses nutrition management among HIV-infected patients in Thailand. Based upon our findings and similar findings in the literature, we make the following recommendations:

- Individualized nutritional counseling should be integrated into routine HIV/AIDS treatment and care.
- Lifestyle modification with aggressive individual nutrition counseling should be in place for 24 weeks in HIV-infected patients with dyslipidemia within ranges specified by NCEP guidelines before initiation of lipid lowering medication.

- Weight control through diet modification should be emphasized in the counseling of individuals with elevated LDL-C.
- Nutritional counseling should encourage reduction in total fat consumption, substitution of saturated fat with unsaturated fat (PUFA and MUFA) and increase in fiber consumption, particularly soluble fiber.
- Knowledge about dyslipidemia should be provided to every HIV-infected patient when they are diagnosed with hyperlipidemia.

Our study has suggested a practical intervention for dyslipidemia management in those receiving antiretroviral therapy with dyslipidemia who had not begun lipid-lowering drug. Nevertheless, future prevention and intervention should also be targeted to HIV patients who are naïve to antiretroviral therapy. If the awareness about dyslipidemia has been raised before initiation of antiretroviral therapy, it may help lower the severity of dyslipidemia in HIV-infected patients receiving antiretroviral therapy. Physicians who care for HIV positive patients should encourage therapeutic lifestyle changes (TLC) in patients before starting lipid lowering medications if patients' lipid levels are still in range where TLC is recommended by the National Cholesterol Education Program (NCEP) guidelines (2002). Furthermore, knowledge about dyslipidemia is one of the factors that predicts reduction of LDL-C by at least 10 mg/dL, therefore HIV infected patients should be provided with knowledge about dyslipidemia and encouraged to attain healthy lifestyles since prior to initiation of antiretroviral therapy.

The strengths of our study are the use of a randomized controlled trial (RCT) and an adequate follow up period to observe changes in lipid levels. The RCT allows us to account for possible differences among the assignment groups with respect to known and unknown factors. Thus,

confounding due to unmeasured factors can be regarded as random. Although we had an approximately 15% dropout in the intervention and the control groups, we found that participants dropped out from the study for reasons unrelated to their treatment assignment or lipid levels and dropout. Therefore, the outcome data in this present study seem to be missing completely at random (MCAR). In addition, we used a mixed model to analyze our data. This analytic strategy can eliminate or reduce bias if data are missing completely at random (Bell, Kenward et al. 2013). In the present study, a few limitations also need to be considered. First, a definite limitation of this study is its low sample size**.** Evaluation of the dietary impact is tempered by the fact that final study size was much lower than planned. This led to low power to detect differences in dietary parameters between the intervention group and the control group. Our study population was low (limited by difficulty in recruitment and widespread use of lipid-lowering medication) and a larger population may be required to demonstrate changes in some parameters. Second, the dietary evaluation was based upon a single 24 hour recall which is notably subject to large intra individual variations in day to day intakes. This may not be sensitive enough to capture the adherence of our dietary intervention. Third, lack of data for soluble fiber and trans fat content in the Thai food database limited our ability to determine the effects of these potential predictors of reduction of LDL-C. Lastly, generalizability of the study is limited. We had a compliant population who were willing to participate the study. The results of this study may not reflect characteristics of those HIV-infected persons who were not interested to participate or the study.

Despite these limitations, this study provides important results for long-term management of HIV-infected patients in Thailand. In addition, it will inform Thai researchers and public health officials who are interested in developing a proper nutritional intervention for their own settings. Further

research should emphasize a larger population and a longer follow up period. A longer follow up period may demonstrate a clearer effect of nutritional intervention on the lipid profile.

References

Bureau of Epidemiology, D. o. D. C., MOPH, Thailand Situation of AIDS patients in Thailand, date 30 September 2012, retrieved on <http://www.boe.moph.go.th/report.php?cat=19&id=1268>.

WHO-SEAR (2012). "Regional Health Sector Strategy on HIV 2011 - 2015."

CDC (2013). “Key Concepts about Measurement Error.”, retrieved on <http://www.cdc.gov/nchs/tutorials/dietary/Advanced/ModelUsualIntake/Info1.htm>.

Bell, M. L., M. G. Kenward, et al. (2013). "Differential dropout and bias in randomised controlled trials: when it matters and when it may not." BMJ 346: e8668.

APPENDIX 1: Risk Assessment Tool for Estimating 10-year Risk of Having a Heart Attack

(NCEP, 2000)

This tool uses information from the Framingham Heart Study to predict a person's chance of having a heart attack in the next 10 years. It is designed to estimate risk in adults aged 20 and older who do not have heart disease or diabetes. Use the calculator below to estimate 10-year risk (see <http://hp2010.nhlbihin.net/atpiii/calculator.asp>).

Age	☐years
Gender	☐Female ☐Male
Total Cholesterol	☐mg/dL
HDL Cholesterol	☐mg/dL
Smoker	☐No ☐Yes
Systolic Blood Pressure	☐mmHg
Currently on any medication to treat high blood pressure	☐No ☐Yes

Calculate 10-year risk

APPENDIX 2.: QUESTIONNAIRES

"Effect of nutritional counseling on low-density lipoprotein (LDL) cholesterol among Thai HIV-infected adults receiving antiretroviral therapy"

QUESTIONNAIRE IDENTIFICATION NUMBER |___|___|___|___|

INTERVIEWER ID |_|_|

DATE OF INTERVIEW: _____/ ____ / _____ (Day/Month/Year)

BLOOD PRESSURE	_____/_____	mmHg
BODY WEIGHT	_____	Kilograms
HEIGHT	_____	Centimeters
WAIST CIRCUMFERENCE	_____	Centimeters
HIP CIRCUMFERENCE	_____	Centimeters

The questionnaire includes the following sections:

Section 0 – Questionnaire identification data	
Section 1 – Background characteristics	10 questions
Section 2 - Risk of cardiovascular diseases	6 questions
Section 3 - Antiretroviral therapy history and route of HIV acquisition	5 questions
Section 4 – Knowledge of hyperlipidemia	10 questions
Section 5 – Alcohol/Physical exercise	8 questions

SECTION 1: BACKGROUND CHARACTERISTICS

Q.No.	Questions Content	Answer Codes		Comment
1.1	What is your gender?	☐ Male	1	
		☐ Female	2	
		Missing	9	
1.2	How old were you at last birthday?	Age in years	[_|_]	
		☐ Don't know	88	
		Missing	99	
1.3	What month and year you were born?	Month	[_|_]	
		☐ Don't know month	88	
		Missing month	99	
		Year	[_|_|_|_]	
		☐ Don't know year	88	
		Missing year	99	
1.4	Where do you live?	☐ Bangkok	01	Thailand
		☐ Nonthaburi	02	has 77
		specify province _____	03-76	provinces
		Missing	99	
1.5	How long have you lived in the city/province specified in question 1.4?	Number of Years	[_|_]	
		☐ Don't know	88	
		Missing	99	

1.6	What is the highest level of school you completed: primary, secondary or higher? SELECT ONLY ONE	☐ Grade 1-6 (Primary)	1
		☐ Grade 7-9 (Secondary: lower level)	2
		☐ Grade 10-12 (Secondary: upper level)	3
		☐ Vocational school	4
		☐ University or higher	5
		Missing	9
1.7	What is your current occupation? SELECT ONLY ONE	☐ Full time: Government	1
		☐ Full time: Private company	2
		☐ Part time: Regular	3
		☐ Part time: Irregular	4
		☐ Owner of business	5
		☐ Student	6
		☐ Unemployed	7
		☐ Other, specify ……	
		Missing	9
1.8	What is your average income per month (Baht)?	☐ 0 - Less than 2,000	1
		☐ 2,000 – 5,000	2
		☐ > 5,000 – 10,000	3
		☐ > 10,000 – 15,000	4
		☐ > 15,000	5
		Missing	9

1.9	What is the current marital status?	☐ Married/ Living as married	1
		☐ Separated/ Divorced	2
		☐ Widowed	3
		☐ Single/ Never married	4
		Missing	9
1.10	With whom do you live?	☐ Parents	1
		☐ Husband/Wife/ Girlfriend/Boyfriend	2
		☐ Siblings/Relatives	3
		☐ Friends/coworkers	4
		☐ Alone	5

SECTION 2: RISK OF CARDIOVASCULAR DISEASES

Q.No.	Questions Content	Answer Codes			Comment
			Yes	No	
2.1	Does anyone in your family have suffered from any of the following problems ?	Diabetes	1	2	If answer No in all problems, skip to Q. 2.3
		Hypertentsion	1	2	
		Cardiovascular diseases (stroke, heart attack, etc.)	1	2	
	INDICATE ALL THAT APPLY	Obesity	1	2	
		Hyperlipidemia	1	2	
2.2	If someone in your family has suffered from problems in question 2.1, how is that person related to you ?	☐ Parents/Sibling	1		
		☐ Uncle/ Aunt/ Grand parents	2		
		☐ Other, specify ……	3		
		Missing	9		
2.3	Have you ever smoked cigarette?	☐ Yes	1		If answer No, skip to Q. 2.5
		☐ No	2		
		Missing	9		

2.4	If you smoked, how often did you smoke during the previous month?	☐ Every day	1
		☐ At least once a week	2
		☐ Less than once a week	3
		☐ Don't know	8
		Missing	9
2.5	Have you ever been told by physician that you have hypertension ?	☐ Yes	1
		☐ No	2
		Missing	9
2.6	Are you currently taking antihypertensive drugs?	☐ Yes	1
		☐ No	2
		Missing	9

SECTION 3 : ANTIRETROVIRAL THERAPY HISTORY AND ROUTE OF HIV ACQUISITION

Q.No.	Questions Content	Answer Codes		Comment
3.1	When was the first time you have known that your HIV testing result is positive?	Specify year ……..	[_|_|_|_]	
		☐ Can't remember	8888	
		Missing	9999	
3.2	What was the route of HIV acquisition in your case?	☐ Heterosexual sex	1	
		☐ Homosexual sex	2	
		☐ Intravenous drug use	3	
		☐ Blood transfusion	4	
		☐ Unknown	8	
		Missing	9	
3.3	When was the first time you have started taking antiretroviral therapy?	Specify month and year	[_|_]/	Q 3.3 – 3.5, data were collected from patients' medical records by PI.
		...../…….	[_|_|_|_]	
		☐ Can't remember	88/8888	
		Missing	99/9999	
3.4	What is the your current antiretroviral therapy ?	Specify …….		
		☐ Don't know	8	
		Missing	9	
3.5	How long have you been taking current antiretroviral therapy ?	Duration in months	[_|_]	
		☐ Don't know	88	
		Missing	99	

SECTION 4 : KNOWLEDGE OF HYPERLIPIDEMIA

Q.NO	Question Content	Answer Codes			Comment
Do you agree with the following statements ?		Yes	No	Don't know	
INDICATE ONE ANSWER PER EACH QUESTION					
4.1	Hyperlipidemia is an increase of the amount of fat such as cholesterol in the blood	1	2	3	
4.2	All cholesterol are bad for health	1	2	3	
4.3	Physical exercise can increase good cholesterol	1	2	3	
4.4	Our body can produce cholesterol from stored carbohydrates and fats	1	2	3	
4.5	There is only one type of lipid in our body that is cholesterol	1	2	3	
4.6	Hyperlipidemia increases risk of heart disease	1	2	3	
4.7	Certain foods such as squids contain high amount of cholesterol	1	2	3	
4.8	Alcohol intake does not relate to hyperlipidemia	1	2	3	
4.9	Hyperlipdemia is always genetic process	1	2	3	
4.10	All patients with hyperlipidemia need to take lipid-lowering medication	1	2	3	

SECTION 5 : ALCOHOL/PHYSICAL EXERCISE

Q.NO	Question Content	Answer Codes		Comment
5.1	In the past 3 months, do you drink alcoholic drink ?	☐ Yes	1	If answer No, skip to Q. 5.4
		☐ No	2	
		Missing	9	
5.2	How often do you drink alcholic drink ?	≤1 drink per month	1	
		2-3 drink per month	2	
		1-2 drink(s) per week	3	
		3-4 drinks per week	4	
		Almost every day	5	
		Daily	6	
		Missing	9	
5.3	Would you say the answer in Q.5.2 was the typical of what you behaved during last month ?	☐ Yes	1	
		☐ No	2	
		Missing	9	
5.4	How much time do you usually spend sitting or reclining per day ? (Do not include time spend for sleeping).	Hours per day	[_|_]	
		Minutes per day	[_|_]	
		Missing	99	

			Yes	No
5.5	During the past 30 days, have you been engaging in any of the following activities ? **INDICATE ALL THAT APPLY**		1	2
		Walking		
		Jogging	1	2
		Bike riding	1	2
		Swimming	1	2
		Aerobics	1	2
		Dancing	1	2
		Calisthenics	1	2
		Gardening	1	2
		Lifting weights	1	2
		☐ Other specify ……		

5.6	During the past 7 days, how many sessions did you engage in vigorous-intensity physical activities∗ for at least 20 minutes at a time?	Session per week	[_]
		☐ Don't know/not sure	8
		Missing	9

5.7	During the past 7 days, how many sessions did you engage in moderate-intensity physical activities∗∗ for at least 20 minutes at a time?	Sessions per week	[_]
		☐ Don't know/not sure	8
		Missing	9

5.8	During the past 7 days, on how many days did you walk for at least 20 minutes at a time ?	Days per week ☐ Don't know/not sure Missing	[_] 8 9

∗*Vigorous activities* are activities that make you breathe much harder than normal and may include heavy lifting, digging, aerobics, or fast bicycling.

∗∗*Moderate physical activities* are activities that make you breathe somewhat harder than normal and may include carrying light loads, bicycling at a regular pace, or doubles tennis. Do not include walking.

APPENDIX 3: FOOD RECORD FORMS

WEEKLY FOOD RECORD FORM

ID ……

Week of …… From Date ……/……/…..…… to Date ……/……/…….. (Day/Month/Year)

	Monday	Tuesday	Wednesday	Thursday	Friday	Saturday	Sunday
Breakfast							
Snack							
Lunch							
Snack							
Dinner							

Notes ……

REVIEWER ID |_|_|

DATE OF REVIEW: _____/ ____ / _____ (Day/Month/Year)

24-HR FOOD RECORD FORM

ID ……

RECORD DATE ……/……/…….. (Day/Month/Year)

Meal / time	Menu	Food details	Rice – starchy food (Rice-serving spoons)	Vegetable (Rice-serving spoons)	Fruit (Portions)	Meat (Spoons)	Milk (Glass)	Oil, sugar and salt (Teaspoon)	Type of oil
Breakfast									
Snack									
Lunch									
Snack									
Dinner									
Snack									
Total									

INTERVIEWER ID |_|_|

DATE OF INERVIEW: _____/ ____ / _____ (Day/Month/Year)